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The Function of Magnesium Compounds in an Oxygen-Alkali-Carbohydrate System

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THE FUNCTION OF MAGNESIUM COMPOUNDS IN AN OXYGEN-ALKALI-CARBOHYDRATE SYSTEM

A thesis submitted by

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SUMMARY

The effects of magnesium ion and other additives on the oxygen-alkali degradation of methyl β-D-glucopyranoside (MBG) were studied in a teflon-lined reactor at 120°C. The glucoside (30-50 mM) in 5% sodium hydroxide solution was treated with oxygen (55-95 p.s.i. equivalent at 25°C.) for up to 13.5 hours.

In the absence of additives, a very short induction period was observed. The reaction produced hydrogen peroxide (H_2O_2) as an intermediate product, as well as other, more stable organic peroxides which were not specifically identified. The identity of the peroxide intermediate was established by comparing it with authentic H_2O_2, in terms of stability in alkali, behavior in acidic titanium sulfate solution, and polarographic response.

Magnesium brought about a lengthening of the induction period, and considerable stabilization of the hydrogen peroxide intermediate. The extended induction period was characterized by a very slow degradation of glucoside, and a fairly constant rate of hydrogen peroxide formation. The rate of H_2O_2 production was equal to the rate of MBG degradation during this induction period. After the induction period, the peroxide level reached a maximum and slowly decreased, while the glucoside degradation attained a faster equilibrium rate which was unaffected by the presence of magnesium compounds.

The addition of ferric ion (0.05 mM $FeCl_3$) caused the level of H_2O_2 to be maintained at or near zero. It also eliminated the induction period, and brought about an increase in the glucoside degradation rate for the first several hours of reaction. Eventually, however, the reaction rate decreased to the same level as that observed without additives.

When iodide ion (10.0 mM KI) was added to the oxygen-alkali-MBG system, complete destruction of the hydrogen peroxide intermediate was effected. This phenomenon was accompanied by an extension of the induction period to over ten hours.

Polarographic work demonstrated that the hydrogen peroxide intermediate produces a double-wave polarogram in 5% sodium hydroxide. The first wave (-0.95 v. vs. S.C.E.) was a catalytic wave which was enhanced by ferric ion, and diminished by a complexing agent and by magnesium ion. The second wave (-1.4 v. vs. S.C.E.) was the normal hydrogen peroxide reduction wave. It was not affected by any of the additives studied.

These observations, along with other evidence, suggested that magnesium ion stabilizes the hydrogen peroxide intermediate by deactivating catalytic metal ions, rather than by interacting directly with peroxide molecules. It was further concluded that this peroxide stabilization by magnesium is directly responsible for delaying the start of the more rapid glucoside degradation. Thus, in the absence of magnesium, trace catalytic ions apparently induce decomposition of the hydrogen peroxide intermediate, to produce species capable of initiating chain reactions (probably hydroxyl and perhydroxyl radicals). The production of these initiating species can be curtailed by substances which deactivate the catalytic ions (such as magnesium), or by those which harmlessly remove the peroxide by an ionic mechanism (such as iodide ion).

A mechanism for glucoside degradation which is consistent with all available data is proposed. It involves the free radical oxidation of secondary hydroxyl groups, to produce alkali-labile keto intermediates. A rate equation derived from this mechanism was capable of predicting MBG degradation rates.

Several experiments were also conducted to investigate the peroxide produced from the reaction of D-glucose in oxygen-alkali. This peroxide also proved to be H_2O_2. It was concluded that a reducing sugar accelerates the oxidation of a nonreducing sugar because of the rapid production of hydrogen peroxide.

INTRODUCTION

SIGNIFICANCE OF THE PROBLEM

The degradation of carbohydrates by oxygen in an alkaline medium is of great practical importance. In the case of polysaccharides such as cellulose, it is manifested primarily by a decrease in viscosity (1). Such degradation is desired in the aging of alkali cellulose. During oxygen bleaching of pulp, however, this type of degradation must be minimized.

Prior to 1964, oxygen bleaching was considered impractical, due to the attendant loss of pulp strength caused by cellulose depolymerization (2-6). Then Robert and associates (6-8) found that, by adding certain inorganic chemicals to the oxygen bleaching system, the mechanical properties of the pulp could be preserved. Notable among these "property protectors" was magnesium carbonate, added at a level of about 1% based on pulp. It has since been demonstrated that other forms of magnesium are also effective in inhibiting carbohydrate degradation (9,10). This discovery of the stabilizing action of magnesium[1] opened the way for the development of oxygen bleaching into an attractive commercial process.

In order to increase the prospects for control of carbohydrate degradation in oxygen-alkali, it is desirable to have an optimum understanding of the chemistry of the system. Of particular interest are the reactions which are affected by such stabilizers as magnesium. The present work was designed to identify such reactions, and to determine how and why they are influenced by magnesium compounds.

[1]The term "magnesium" will sometimes be used to designate magnesium ion or any of several magnesium compounds. It does not refer to elemental magnesium metal.

LITERATURE REVIEW

INTRODUCTORY COMMENT

The purpose of this review is to acquaint the reader with the historical foundation upon which this thesis was developed. For further detail, the literature may be consulted. Of particular interest are the surveys presented by McCloskey (11) and by Norin (12).

OXIDATION UNDER OXYGEN BLEACHING CONDITIONS

Oxygen bleaching is normally carried out in 0.5-5.0% sodium hydroxide under 5-15 atmospheres oxygen pressure. Reactions are conducted at 90-130°C. for less than two hours. Pulp consistencies are generally in the vicinity of 20% (8).

A major cellulose reaction undoubtedly involves attack at C2 or C3, to produce alkali-labile carbonyl groups (13-16). Theander has established that 2- and 3-keto derivatives of MBG (17,18) and cellulose (19) are extremely sensitive to alkali. Such alkaline degradations of carbonyl derivatives are believed to occur via β-alkoxy elimination, to yield aldonic acids (14,15,20). Samuelson and coworkers found that oxygen-alkali cleavage of glycosidic bonds in cellulose (12,13,18) and xylan (16) produced a variety of aldonic acid end groups. In their studies with MBG, however, Ericsson, et al. (21) and McCloskey (11) found that this model compound produced some acidic products wherein the "glycosidic bond" was not cleaved.

Peroxides were detected as intermediates in reactions of cellulose (22), cellobiitol (11,13), 1,5-anhydrocellobiitol (11), and MBG (11). Model compounds which had a greater number of available hydroxyl groups produced higher concentrations of peroxides (11).

The presence of easily oxidizable organic compounds such as glucose or lignin caused an increase in the degradation of MBG (11), and increased the oxygen consumption by cellobiitol (13). This accelerated oxidation was accompanied by an increase in peroxide production. Carbohydrate degradation was also markedly enhanced by the addition of a few parts per million of transition metal ions. The metal ions, however, brought about a decrease in the concentration of the peroxide intermediate (11,13).

Samuelson and Stolpe (13) indirectly verified the stabilizing effects of magnesium carbonate, using cellobiitol as a model compound. Although these workers reported no carbohydrate analyses, they did establish that the magnesium compound diminished oxygen consumption and organic acid production from cellobiitol, while increasing the maximum peroxide concentration. Magnesium had the ability to counteract the accelerating effects of glucose, lignin, and transition metal ions. McCloskey (11), on the other hand, found that magnesium carbonate exerted only minimal effects on the degradation of 10 mM MBG in oxygen-alkali.

From his kinetic study of MBG degradation, McCloskey (11) concluded that the reaction was second-order in glucoside, first-order in oxygen pressure, and first-order or variable in alkali concentration. He obtained most of his data at a glucoside concentration of 10 mM.

ALKALI CELLULOSE AGING

One of the steps in the manufacture of viscose rayon involves the aging, or autoxidation, of alkali cellulose. The objective of aging is to reduce the viscosity of cellulose prior to xanthation. A typical treatment would involve steeping cellulose, at ca. 30% consistency, in 15% sodium hydroxide at 25-40°C. The reaction is allowed to proceed for several hours, in contact with air (23).

Although the reaction conditions used in aging and in oxygen bleaching are quite different, several workers (13,20,24) have noted many similarities between the two processes. Indeed, it would not be surprising to find that the carbohydrate chemistry was identical in the two cases, since the same reactants are present.

The mechanism of the aging reaction was the subject of papers by Entwistle, et al. (25), Mattor (26), and MacDonald (27). The latter worker (27) combined ideas of his predecessors (25,26), and proposed a mechanism involving both ionic and free radical intermediates. He suggested that the reaction started after hydrogen peroxide was produced from carbohydrate reducing groups. Complexes between the peroxide and transition metal impurities then supposedly reacted with cellulose, to produce chain-initiating free radicals. Cellulose hydroperoxides were among the postulated species involved in chain propagation. MacDonald (27,28) also established that sulfur inhibits the aging reaction. This was cited as evidence that free radicals are indeed involved.

The above mechanisms (25-27) require oxidation of a reducing end as an initial reaction. Michie and Neale (29), however, advanced the opinion that the reaction starts by oxidation of a hydroxyl group along the chain. Clearly a reducing group could not be the exclusive site for an initial reaction in oxygen-alkali, since nonreducing sugars such as MBG do react (11).

Michie and Neale (29) also conducted a kinetic study of the aging reaction. They found the cellulose degradation rate to be between first- and second-order in cellulose, about first-order in hydroxyl ion concentration, and between zero- and first-order in oxygen pressure. There was also an undefined dependency on the metal ion content of the system.

OXYGEN-ALKALI REACTIONS OF REDUCING SUGARS

Since we are ultimately interested in polysaccharides which contain both reducing and nonreducing functions, it is fitting to consider briefly oxygen-alkali reactions of reducing sugars. As mentioned above, the presence of a reducing sugar profoundly influences the degradation of a nonreducing sugar (11,13).

In alkaline solution, oxygen very readily degrades reducing sugars to give high yields of acids having one carbon atom less than the sugar (30-35). In excess oxygen and alkali, the production of such C1-C2 cleavage products is often practically quantitative (36,37).

Bamford and Collins (36) studied oxidation of D-glucose at room temperature. They noted that peroxides were formed initially at the same rate as oxygen was consumed. The reaction was not catalyzed by most metal ions, nor inhibited by free radical scavengers.

Malinen and Sjostrom (35) investigated the oxygen-alkali reactions of cellobiose at 120°C. This reaction was much more complex than that at room temperature. The authors noted catalysis by metal ions, and inhibition by magnesium compounds, at the higher temperature.

OBJECTIVES OF THE THESIS

The primary objective of this investigation was to study the mechanism by which magnesium compounds inhibit carbohydrate degradation in an oxygen-alkali medium. A secondary objective, related to the first, was to obtain information regarding the identity and role of the peroxide intermediate. It was also desired that the thesis might improve our understanding of the carbohydrate degradation reactions in such a system.

ALTERNATIVE HYPOTHESES

INTRODUCTORY COMMENT

In the light of the literature review, several hypotheses can be advanced to explain the stabilizing action of magnesium compounds. These are illustrated graphically in Fig. 1. A brief discussion of each hypothesis follows.

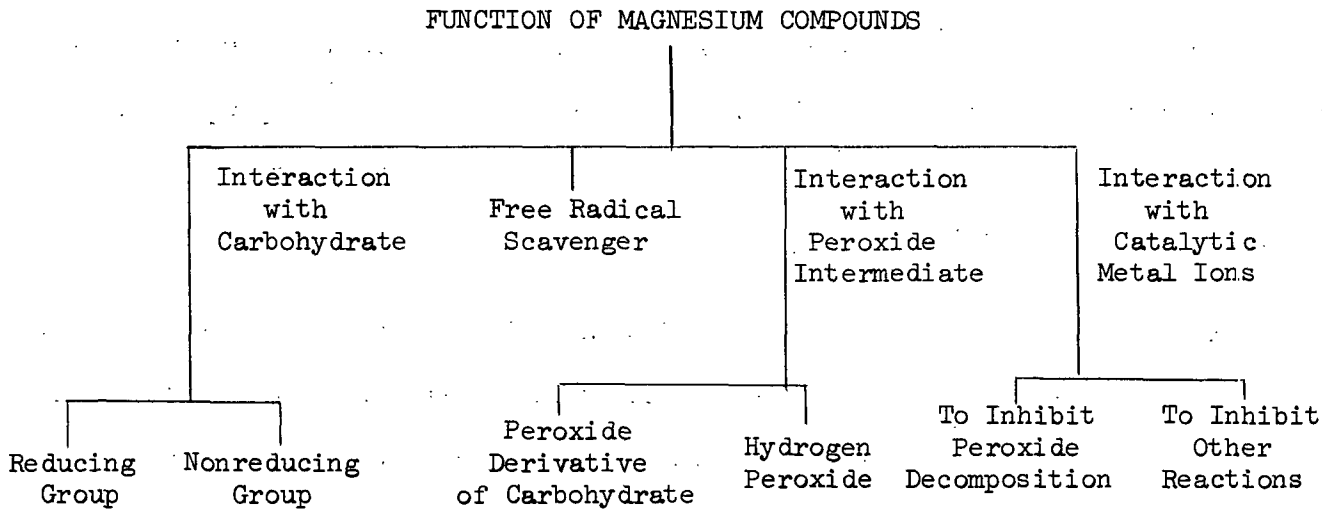


Figure 1. Alternative Hypotheses Regarding Stabilization of Carbohydrate in Oxygen-Alkali by Magnesium Compounds

INTERACTION WITH CARBOHYDRATE

Magnesium might prevent oxidation at an active location on a carbohydrate molecule by forming a complex or bond at that site. This location could be an aldehyde function, carboxyl group, or other anion at a reducing end, or carbonyl and/or hydroxyl groups along the chain. The reader will recall that magnesium was not very effective in stabilizing 10 mM MBG (11). This is consistent with the hypothesis that magnesium interacts primarily with reducing groups, or some other function not found in the model compound.

FREE RADICAL SCAVENGER

If the reaction involved free radical intermediates, magnesium could inhibit it by acting as a radical scavenger. Such inhibition would be manifested by a slower degradation rate throughout the entire reaction period.

INTERACTION WITH PEROXIDE INTERMEDIATE

Regardless of whether the peroxide intermediate is an organic peroxide or H_2O_2, it is conceivable that magnesium could form a complex with it. Such complexing could inhibit some subsequent reaction of the peroxide which is involved in carbohydrate degradation. This subsequent reaction might involve direct attack of the carbohydrate by the peroxide, or peroxide decomposition to yield other active species, such as radicals.

Samuelson and Stolpe (13) established that magnesium ion stabilized the peroxide produced from cellobiitol, while it diminished oxygen consumption and organic acid production. Magnesium is also known to stabilize alkaline hydrogen peroxide (38). It is apparently not known, however, if magnesium ever forms a direct complex with any peroxide. Nor has a direct connection been established between the peroxide-stabilizing ability of magnesium, and its inhibition of carbohydrate degradation.

INTERACTION WITH CATALYTIC METAL IONS

Multivalent metal ions generally induce decomposition of peroxides by a free radical mechanism (38-40). Perhaps even minute traces of transition metal ions are sufficient to produce significant amounts of chain-initiating radicals or other active species from the peroxide intermediate. Magnesium could minimize the production of these active species by complexing with or deactivating the catalytic ions.

Multivalent metal ions could also hypothetically produce radicals by reacting with species other than the peroxide, as in Reactions (1) and (2):

$$RO^- + M^{+n+1} \longrightarrow RO\cdot + M^{+n}, \qquad (1)$$

$$O_2 + M^{+n} \longrightarrow O_2^- + M^{+n+1}, \qquad (2)$$

where RO^- represents an ionized hydroxyl group of a carbohydrate molecule, and M represents a transition metal. Magnesium could also inhibit Reactions (1) and (2) by interaction with the metal ions.

EXPERIMENTAL APPROACH

A model reaction system was used to study the effects of magnesium compounds, and other additives and experimental variables, on carbohydrate degradation under oxygen bleaching conditions. MBG was chosen as a model compound, because it is readily available in pure form, it is soluble in alkaline solutions, and its alkaline hydrolysis in the absence of oxygen is insignificant at the 120°C. temperature used in this work (11). Reaction conditions were similar to those used by McCloskey (11), except that higher glucoside concentrations were employed. This allowed significant carbohydrate degradation to be observed after a shorter reaction time. Several experiments were also conducted with D-glucose as a model reducing sugar. The teflon-lined reactor, described in the following section and in Appendix I, was of unique design. It permitted periodic sampling of the hot reaction solutions under pressure, for peroxide and sugar analysis.

As in any model compound study, the question of the suitability of the model invariably arises. The chemical and physical differences between cellulose and MBG are obvious. In addition, the presence of lignin, hemicellulose, and other components of pulp introduce complicating factors not found in the model system

(24). Moreover, MBG was allowed to react to an extent of about 20-30% in the present work, whereas an oxygen bleach is completed long before such extensive degradation occurs. Nevertheless, the author believes that many of the conclusions drawn from this investigation can be applied to cellulose degradation and stabilization, during both oxygen bleaching and alkali cellulose aging.

In addition to the carbohydrate reactions, experiments were conducted to compare the chemical properties of the peroxide intermediate with those of hydrogen peroxide. The stabilities of the peroxides in alkali were investigated, along with their polarographic behavior. Polarography also proved to be an extremely useful and profitable tool in studying interactions between the peroxides, magnesium ion, and transition metal ions.

EXPERIMENTAL

OXYGEN-ALKALI REACTIONS

Details regarding the preparation and purification of MBG, water, sodium hydroxide, and other reagents, are given in Appendix II. A brief description of the construction and use of the reactor is presented in this section. A more thorough discussion is given in Appendix I.

Solutions of MBG (30-50 mM) in 1.25N alkali were prepared by dissolving appropriate amounts of the glucoside, stock sodium hydroxide solution, and any other necessary reagents, in triply-distilled water (usually 250 ml.). Immediately after preparation, the solution was transferred to the magnetically-stirred teflon-lined stainless steel reactor (volume ca. 800 ml.). After sealing and purging with nitrogen, the reactor was immersed in a preheated oil bath. A period of at least 90 minutes was allowed for the reactor and contents to attain thermal equilibrium at 120°C. The reaction was then started by pressurizing to the desired level with oxygen. Although the oxygen was added to the system at 120°C., pressures are reported herein as equivalent partial pressures of oxygen at 25°C.

The reactor cover was equipped with two ports. Through one port, a length of teflon spaghetti tubing extended from the solution in the reactor, through a cooling coil, to a system of valves. This valve system was used for adding unstable reagents (such as H_2O_2) after heat-up, and for sampling the solution at any desired reaction time. Through the other port, the space above the reaction solution was connected to a pressure gage and the oxygen supply. The oxygen line was kept open throughout the reaction. This allowed a constant oxygen pressure to be maintained above the solution as oxygen was consumed and as samples were withdrawn.

Oxidations of glucose were carried out in the same reactor at room temperature. In this case, only sodium hydroxide and water were put into the reactor prior to sealing and heat-up. The glucose was dissolved in 2 ml. of water, and forced into the reactor with oxygen at the start of reaction.

ANALYSIS OF REACTION SOLUTIONS

Analytical methods employed in this study included determinations of peroxides and unreacted glucoside. Detailed descriptions of the procedures may be found in Appendix III.

The concentrations of peroxide intermediate produced from MBG and glucose were determined by colorimetric analysis of a yellow complex between titanium ion and the peroxide in sulfuric acid. In the case of MBG reaction samples, the absorbance of these colored solutions frequently increased upon standing for several hours. As discussed below, this was apparently caused by the presence of other more stable peroxides produced from MBG. The concentrations of these "other peroxides" were estimated from the magnitude of the absorbance increase.

Prior to analysis for unreacted glucoside, samples were deionized with cation exchange and mixed bed resins. This procedure left only unreacted starting material in solution (11). Glucoside determinations were then conducted, using a colorimetric method which employs phenol in sulfuric acid.

PEROXIDE STABILITY STUDIES

An experiment was conducted to compare the stability of the peroxide intermediate from MBG with that of H_2O_2 in the same medium. MBG (50 mM) was reacted in 1.25N sodium hydroxide under 95 p.s.i. oxygen at 120°C. for 1.6 hours. An aliquot of this reaction solution (30 ml.) was made 1.0 mM in magnesium sulfate,

and placed in a pyrex vessel in a 35°C. water bath. Portions of the treated aliquot were withdrawn at intervals, and analyzed for peroxide by the titanium method. The remainder of the original untreated reaction solution was transferred to a pyrex vessel and held at room temperature for one week, at which time the peroxide in the sample had all decomposed. It was then made 1.0 mM in magnesium sulfate, and hydrogen peroxide was added. The stability of the H_2O_2 in this solution at 35°C. was likewise determined by analyzing for peroxide at several time intervals.

The room temperature decomposition of hydrogen peroxide in 1.25N sodium hydroxide in the presence of magnesium sulfate was investigated. The solutions were prepared by dissolving magnesium sulfate in water contained in a 50-ml. volumetric flask, adding sodium hydroxide, then peroxide, and diluting to the mark. The titanium method was again employed on samples taken at intervals.

The stability of alkaline H_2O_2 at 120°C. in the presence of various additives was also studied. These experiments were conducted by using the reactor, as described above. All reagents but peroxide were put into the reactor prior to sealing. Hydrogen peroxide was forced in, after heat-up, at zero reaction time. Oxygen or nitrogen was added to the equivalent of 75 p.s.i. at 25°C.

Peroxide stability results are reported as decomposition constants. These were obtained as the slopes of plots of log peroxide concentration vs. time.

POLAROGRAPHY

Polarograms were obtained using a Sargent Model XV Recording Polarograph. Unless otherwise noted, polarograms were run at 20°C., with a mercury column height of 60.8 cm., in 1.25N sodium hydroxide as supporting electrolyte. Under these conditions, the capillary characteristics were such that the mercury flow

rate ($\underline{m}$) was 1.298 mg./sec., and the average drop time ($\underline{t}$) was 5.02 sec. in the applied potential range of -1.0 to -1.8 volts. The resistance of the 40-ml. capacity cell was determined to be less than 600 ohms. A further discussion of the apparatus and its use is presented in Appendix I.

RESULTS AND DISCUSSION

OXYGEN-ALKALI REACTIONS OF METHYL β-GLUCOSIDE

PEROXIDES IN REACTION SOLUTIONS

This section includes a discussion of peroxides produced during the oxygen-alkali reaction of MBG. In subsequent sections, the effects of various additives on these peroxides, and on the MBG degradation rate, will be considered.

The acidic titanium sulfate method used for determining peroxide concentrations in oxygen-alkali-MBG reaction samples is described in Appendix III. Modifications of this procedure have been used by other workers (41-46) for quantitative analysis of a variety of organic peroxides, as well as hydrogen peroxide.

In the present study, when pure H_2O_2 solutions were made about 1N in sulfuric acid, a yellow color developed immediately upon adding titanium sulfate. The color intensity remained unchanged upon standing for about two days. After this the colorimetric absorbance gradually diminished. When this colorimetric analysis was applied to acidified MBG reaction samples, color also developed immediately upon adding the titanium reagent. However, the absorbance of these test solutions frequently increased upon standing for several hours. The maximum absorbance was usually attained after about two days. The absorbance started to decrease after this.

The initial absorbance of MBG test solutions corresponded to a peroxide content which was essentially the same as that obtained when the samples were analyzed by a thiosulfate titration (procedure Appendix III).

The subsequent absorbance increase of MBG test solutions was unaffected by nitrogen purging while standing. Thus, it was evidently not caused by new peroxides formed by the action of dissolved oxygen. In one instance, a sample which had attained its maximum absorbance was treated with potassium fluoride to release peroxide from the titanium complex (41). A subsequent titration with thiosulfate revealed the same peroxide level as that corresponding to the maximum absorbance. Hence, all the color in the test solution was indeed due to the complexing of peroxide in the sample.

The titanium reagent forms the colored complex with the O_2^{-2} species (41-46). Hydrogen peroxide reacts immediately, whereas organic peroxides react as they are hydrolyzed to hydrogen peroxide in the strong acid. The basis for distinguishing between these peroxides lies in differences in the rates of their hydrolysis to H_2O_2. Therefore, the absorbance increase encountered here was apparently caused by slow hydrolysis of organic peroxides in the sample. Since these other peroxides may undergo decomposition reactions other than hydrolysis during the two days' standing period, the absorbance increase probably provides a low estimate of their concentration. This is apparently the first report of the formation of peroxides other than the "peroxide intermediate" in an oxygen-alkali-carbohydrate system.

Since the identity of the organic peroxides which may be in a sample is unknown, it is uncertain as to how readily they may be hydrolyzed to H_2O_2 in the acidic test solution. Conceivably, part of the initial absorbance could be due to organic peroxides which are immediately converted to H_2O_2 in the presence of acid. For example, α-hydroxy hydroperoxides (40,46) undergo the rapid equilibrium

$$R_1R_2C(OH)OOH \rightleftharpoons R_1R_2CO + H_2O_2 \ . \qquad (3)$$

It is doubtful, however, that such organic peroxides would be stable under the alkaline reaction conditions employed here (40).

Marklund (47) was able to use a titanium method to distinguish α-hydroxy peroxides and hydroperoxides from H_2O_2. A distinguishing feature of his procedure involved the use of less concentrated acid in the test solution. Thus, when the test was carried out at pH 1 or above, H_2O_2 reacted immediately, but hydroxymethyl hydroperoxide ($HOCH_2OOH$) required several minutes to attain complete color development. Increasing the pH slowed down the hydrolysis rate.

Varying the pH of the test solution from 0 to 3 in the present work did not affect the color response of pure H_2O_2. When a mixture of cyclohexanone peroxides (obtained from Dr. N. S. Thompson) was analyzed at the higher pH, an immediate color response was observed (H_2O_2), followed by a gradual absorbance increase (organic peroxides). When MBG reaction samples were analyzed, however, the same immediate color response was observed in the entire pH range of 0 to 3. This was taken as evidence that H_2O_2 was the only peroxide causing the initial color response.

Increasing the pH did slow down the hydrolysis rate of the "other peroxides" in the MBG samples. No absorbance increase was observed after two days at pH 1 and above, and only a small increase in color was exhibited at pH 0.5.

EFFECTS OF MAGNESIUM COMPOUNDS

MBG was oxidized in the presence and absence of magnesium oxide; glucoside analyses are presented in Fig. 2. Whenever duplicate runs were made, the average results were plotted. In such cases, standard deviations of the averages were less than 2%. Data for these and other reactions are listed in Table IV of Appendix IV. Reaction rates are tabulated in Table V.

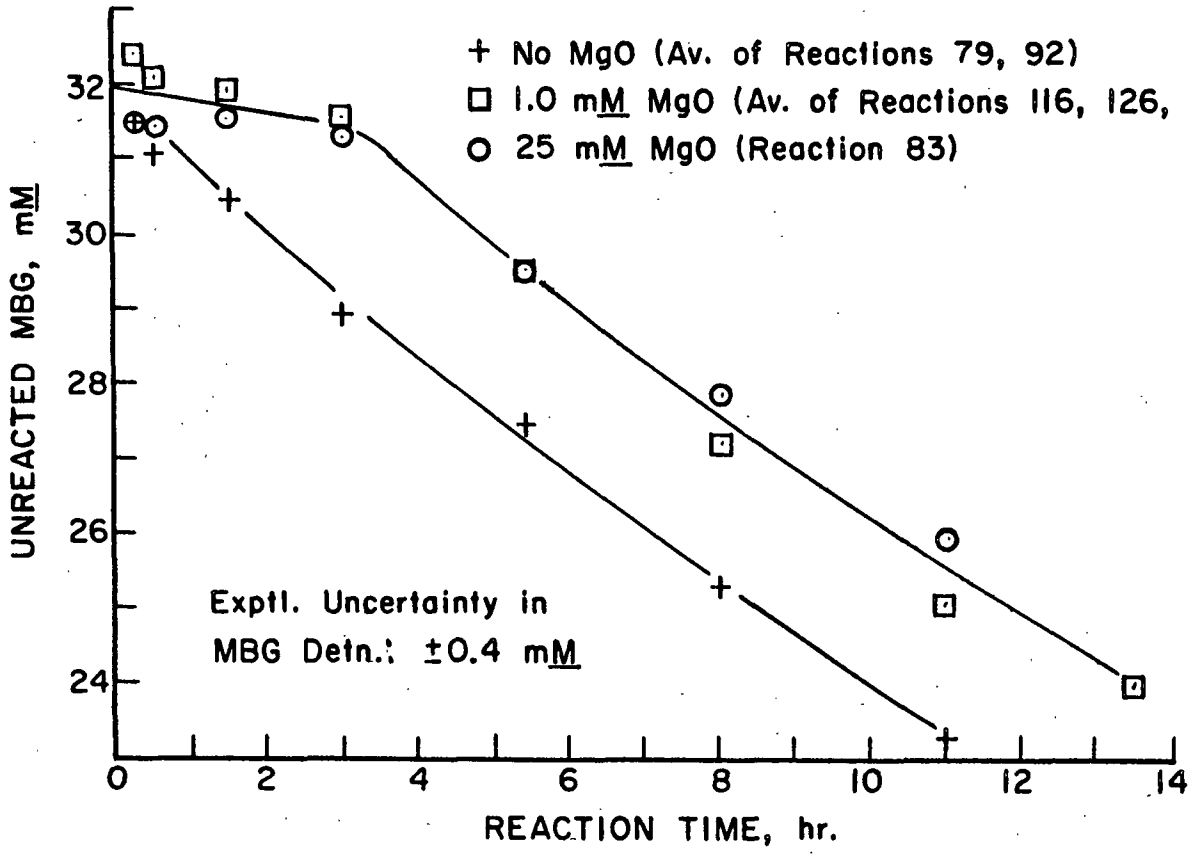


Figure 2. Effect of MgO on the Degradation of MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2

The primary effect of magnesium on MBG oxidation involved the lengthening of an induction period. Although Minor and Sanyer (48) observed an induction period in the oxygen-alkali reaction of D-glucitol, this is apparently the first report of a connection between the length of such an induction period and the presence of magnesium compounds. Magnesium produced no detectible change in the MBG degradation rate after the initial induction period.

When 1.0 mM magnesium oxide was used, no precipitate was observed in the solutions. When 25 mM magnesium oxide was added, however, a large portion of it

did not dissolve. Nevertheless, these experiments with the two magnesium concentrations gave glucoside results which could not be declared significantly different from each other (Fig. 2). Although the higher magnesium charge may have exerted a slight stabilizing effect at longer reaction times, the difference was still within experimental uncertainty. An experimental error of about ± 0.4 mM was associated with the determination of unreacted glucoside at a given reaction time.

During the induction period, there appeared to be a slow degradation of MBG. The experimentally observed rate in this case was between 0 and 0.2 mM/hr.

Figures 3 and 4 illustrate the effects of magnesium on the peroxides produced from MBG. The concentrations of the "peroxide intermediate" and "other peroxides" were determined as discussed above. Reaction solutions which contained the magnesium precipitate were analyzed both before and after allowing the precipitate to settle out. Removal of the precipitate did not affect the peroxide results, indicating that all the peroxide was soluble.

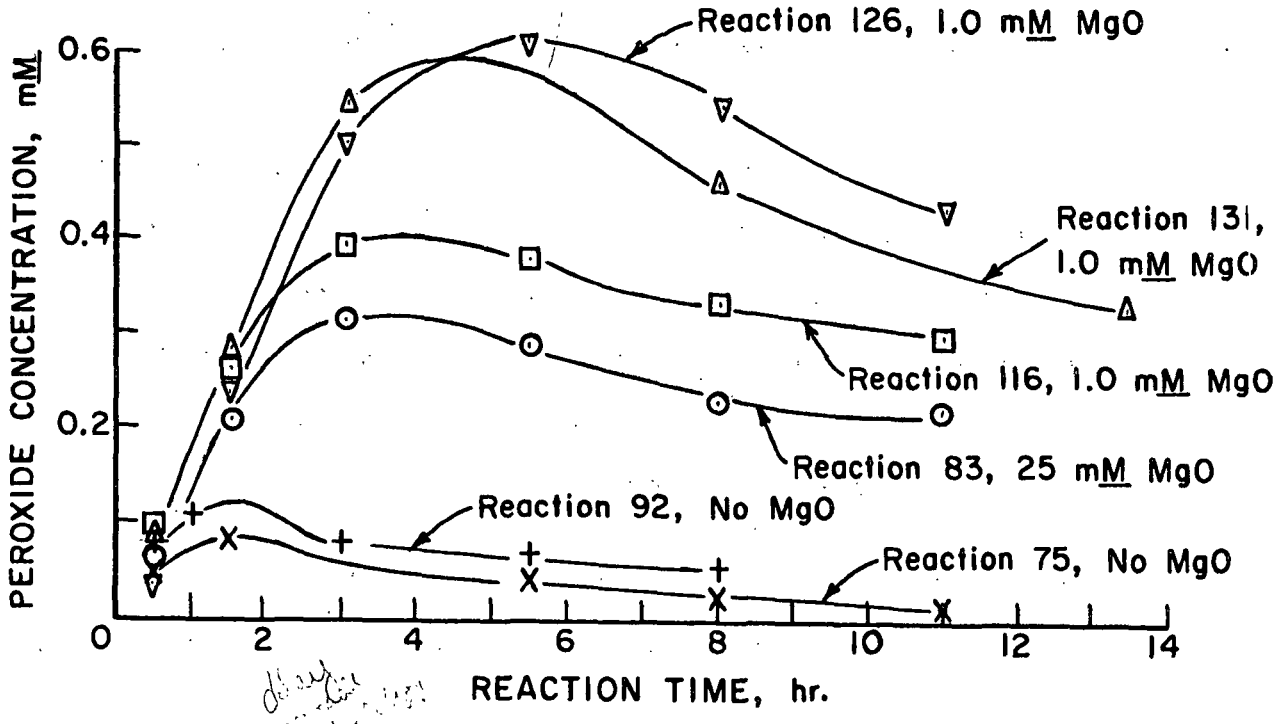


Figure 3. Production of "Peroxide Intermediate" from 30 mM MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2

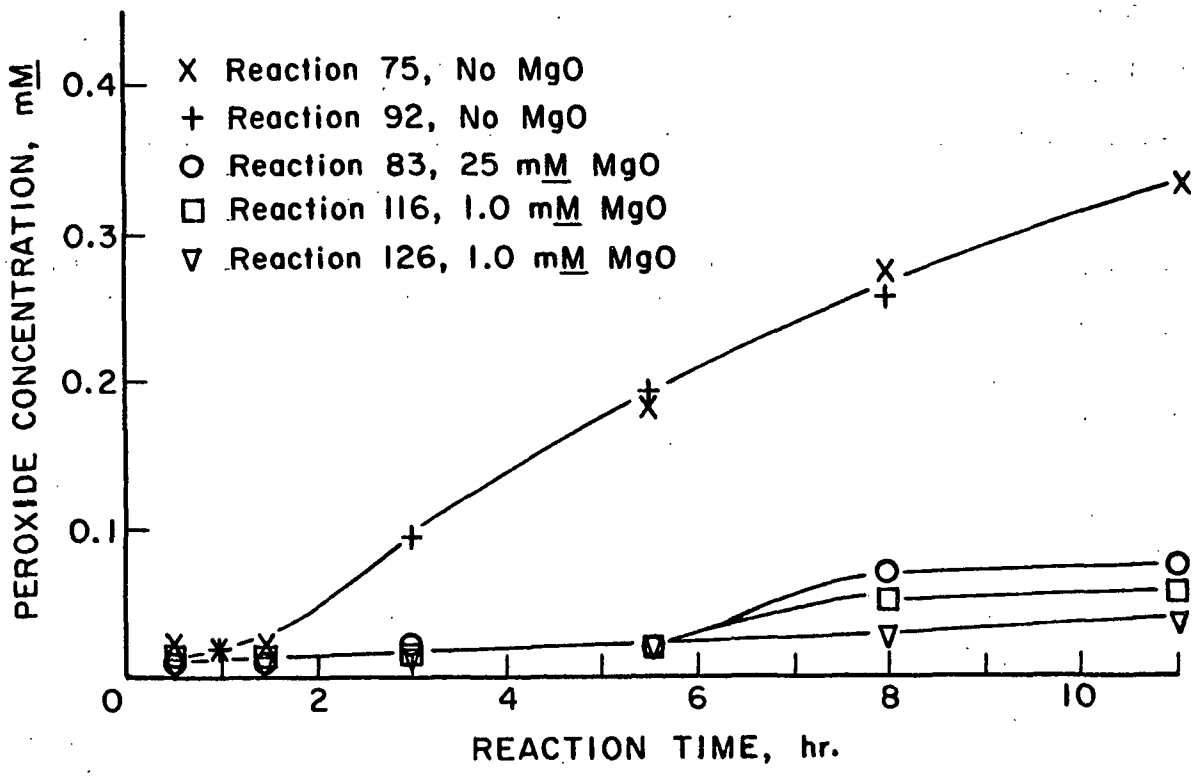


Figure 4. Production of "Other Peroxides" from 30 mM MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2

Magnesium caused an increase in the maximum concentration of the hydrogen peroxide intermediate. The results with cellobiitol reported by Samuelson and Stolpe (13) are in agreement with this. The reason for the discrepancy between the supposedly duplicate Reactions (116), (126), and (131) (Fig. 3) is uncertain. It may, however, be due to unavoidable trace contaminants in the solutions. This point is discussed further in a later section.

Magnesium appeared to have no influence on the rate of accumulation of the peroxide intermediate. Rather, its effect seemed to be related to delaying the time at which peroxide decomposition became important. When magnesium delayed the start of peroxide depletion, it also delayed the start of the more rapid glucoside degradation. This suggests a correlation between H_2O_2 decomposition and MBG degradation.

In the presence of magnesium, the concentration of the hydrogen peroxide intermediate initially increased at the rate of 0.15-0.20 mM/hr. The similarity between this rate, and the rate of MBG degradation during the induction period mentioned above, is probably not coincidental. It implies that, during the induction period, MBG may have been slowly reacting with oxygen to produce an equivalent amount of the peroxide.

Magnesium caused less of the "other peroxides" to be detected in the reaction solutions. As shown in Fig. 4, higher maximum peroxide intermediate concentrations were accompanied by lower concentrations of the other peroxides.

EFFECTS OF FERRIC AND IODIDE IONS

McCloskey (11) reported that MBG degradation was faster when reactions were run in unlined stainless steel vessels. Similar accelerative effects were noted in preliminary experiments here, when metal ions were accidentally introduced into reaction solutions.

When 0.05 mM ferric chloride was added to the MBG system, the initial degradation rate was significantly increased (Fig. 5). These effects were only temporary, however. The reaction rate after about two hours, in the presence of added ferric ion, was essentially the same as that in the absence of additives. This amount of ferric ion was sufficient to practically eliminate the peroxide intermediate (Fig. 6). It did, however, bring about a slight increase in the amount of other peroxides detected.

It is noteworthy that the level of the peroxide intermediate at extended reaction times had little influence on the MBG degradation rate after the induction period. As shown in Fig. 2, 3, 5, and 6, essentially the same rate was observed after the induction period, whether the peroxide level was high (in the

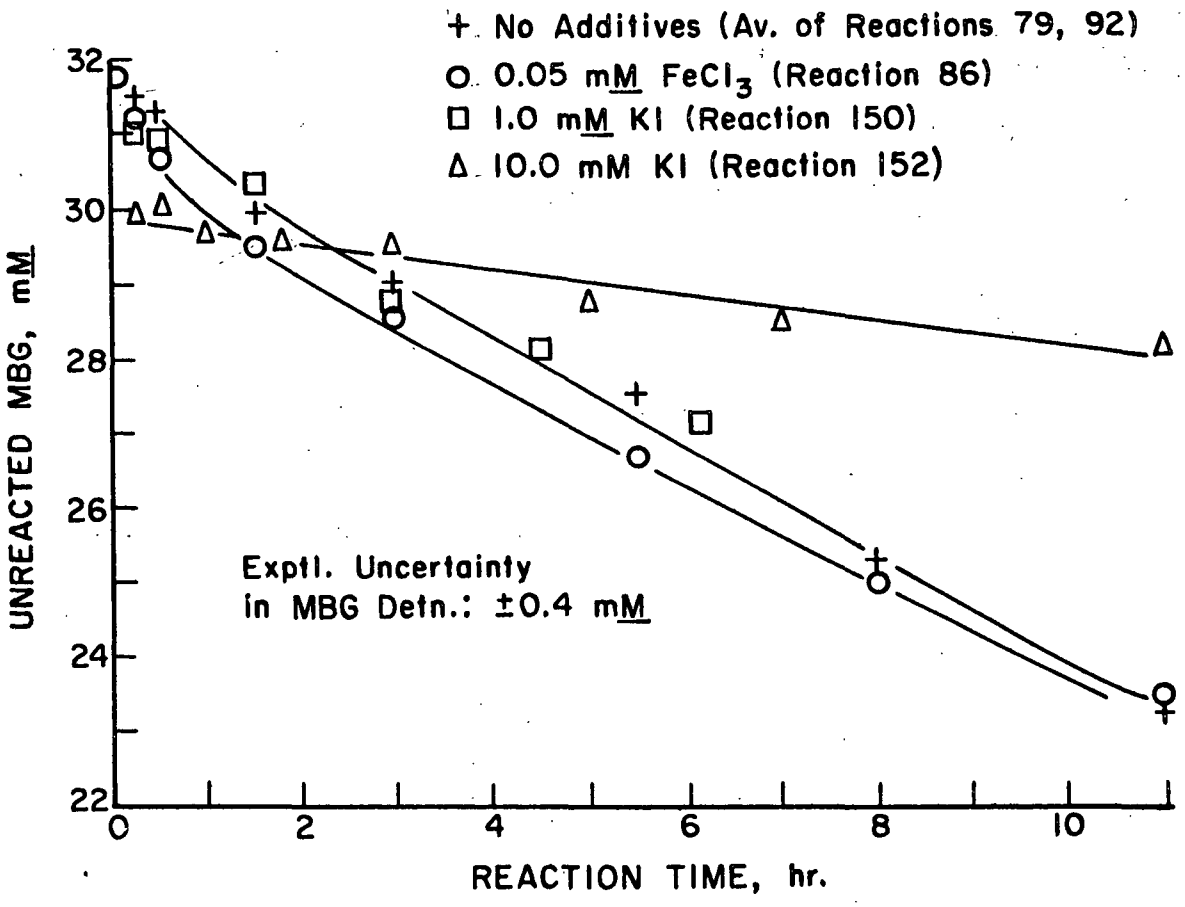


Figure 5. Effects of Ferric and Iodide Ions on the Degradation of MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2

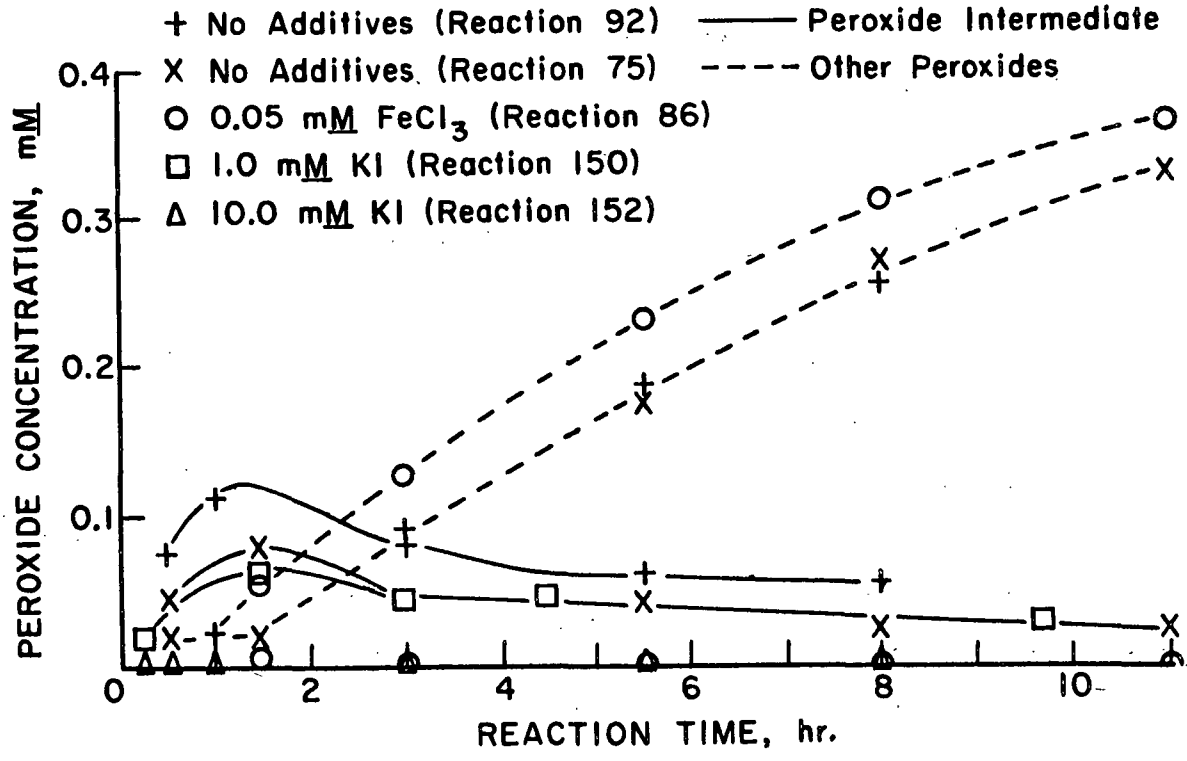


Figure 6. Effects of Ferric and Iodide Ions on the Peroxides Produced from MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2

presence of magnesium), or very low (in the presence of added ferric ion). This independence of final rate on the concentration of the peroxide intermediate implies that the reaction does not involve a direct attack on the glucoside by the peroxide.

Ferric and other transition metal ions generally react with peroxides to produce free radical intermediates (38-40), which, as mentioned previously, may be responsible for initiating glucoside degradation. The iodide-iodine couple, on the other hand, destroys peroxides by an ionic oxidation-reduction mechanism. Reactions (4) through (7) are believed to occur in the case of hydrogen peroxide in alkali (39,49):

$$2I^- + H_2O_2 \longrightarrow I_2 + 2OH^- \qquad (4)$$

$$I_2 + 2OH^- \rightleftharpoons OI^- + I^- + H_2O \qquad (5)$$

$$I_2 + OOH^- \longrightarrow 2I^- + O_2 \ (+ H^+) \qquad (6)$$

$$3OI^- \longrightarrow IO_3^- + 2I^- \qquad (7)$$

Thus, if iodide ion were added to the MBG reaction system, it might harmlessly destroy the peroxide intermediate as it was formed, without producing radicals or other reactive species.

The effects of potassium iodide (1.0 and 10.0 mM) on the MBG reaction are illustrated in Fig. 5 and 6. The analysis for "other peroxides" could not be carried out in the presence of iodide ion. This was due to interference caused by the slow conversion of iodide to iodine in the acid test solutions.

The presence of 1.0 mM iodide ion only slightly affected the MBG reaction. The peroxide-iodide interaction was probably too slow to be important under these conditions. It is known that Reaction (4) is extremely slow in alkali (39).

Increasing the iodide concentration to 10.0 mM did, however, effect complete peroxide intermediate destruction. This was accompanied by a carbohydrate stabilization which was even more pronounced than that caused by magnesium. The induction period was extended indefinitely, so that the MBG degradation rate was only 0.15-0.20 mM/hr. for the entire ten hours' reaction. Again, the reaction rate during this extended induction period was the same as the initial rate of peroxide production observed in the presence of magnesium.

These results are in agreement with those reported by Minor and Sanyer (48). They found that iodide ion stabilized cellulose and D-glucitol in oxygen-alkali. They did not, however, demonstrate that the peroxide intermediate was simultaneously destroyed.

Iodide ion was apparently not acting as a radical scavenger in the present work. The postinduction period degradation rate was not changed by the presence of 1.0 mM KI; a radical scavenger would be expected to decrease this rate. Rather, the stabilization by iodide appeared to be connected with its ability to remove the peroxide intermediate before it could react by a different pathway. These results, in connection with the previously noted effects of ferric ion, are consistent with the idea that the more rapid MBG degradation is initiated by a catalyzed free radical decomposition of the peroxide intermediate.

EFFECTS OF PYROPHOSPHATE AND OXALATE

If the initiation of the more rapid glucoside degradation were caused by a metal ion-peroxide interaction, it might be possible to stabilize carbohydrate by the addition of metal ion-complexing agents. Two such complexing agents were investigated here: potassium oxalate, and the well-known stabilizer for hydrogen peroxide, sodium pyrophosphate (38,39).

As shown in Fig. 7, sodium pyrophosphate had no significant effect on MBG degradation. Nor were the peroxide levels changed much by the presence of this additive (Fig. 8). It is possible that pyrophosphate may have undergone hydrolysis and/or oxidation under the conditions of reaction, or it may simply be ineffective as a complexing agent in alkali.

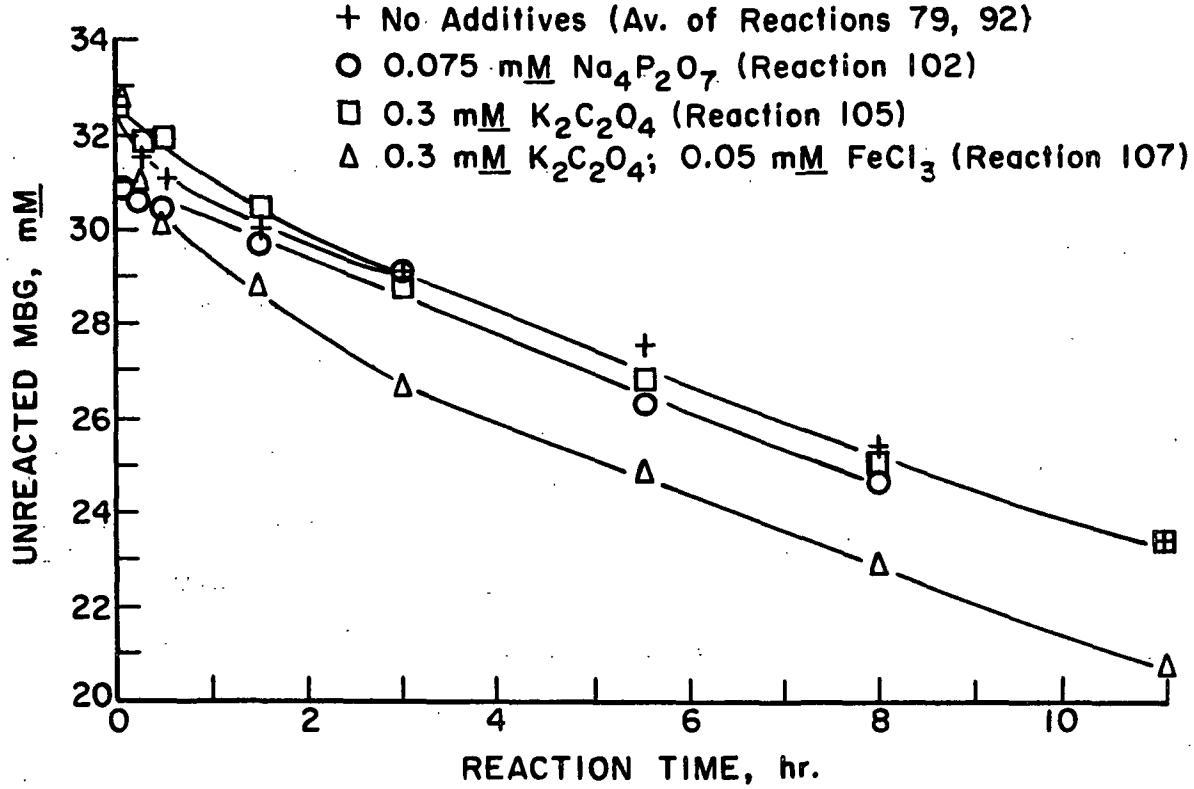


Figure 7. Effects of Pyrophosphate and Oxalate on the Degradation of MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2

A similar lack of effectiveness was exhibited by potassium oxalate. This additive did not affect peroxide levels much, and glucoside degradation was not inhibited. In fact, the initial MBG degradation rate seemed to be increased by the addition of oxalate [Reaction (105), Fig. 7].

Martell and Calvin (50) have discussed a mechanism whereby oxalate chelates may decompose to produce free radical intermediates:

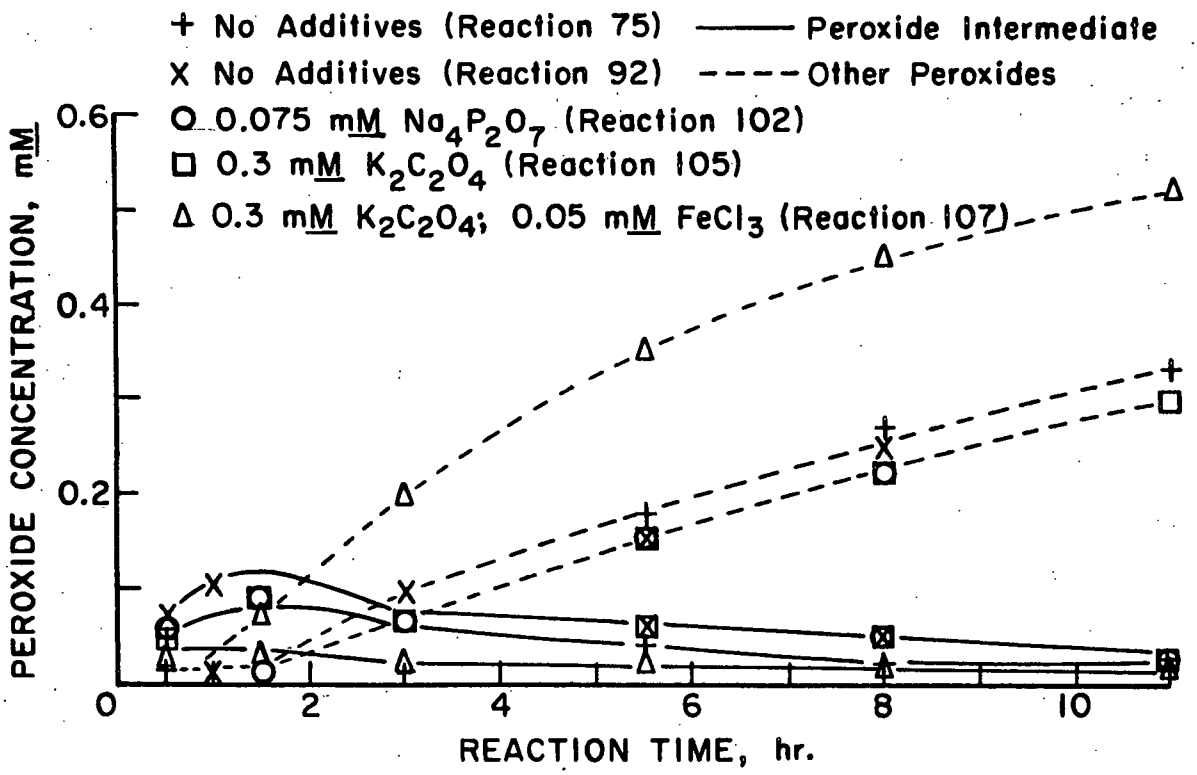
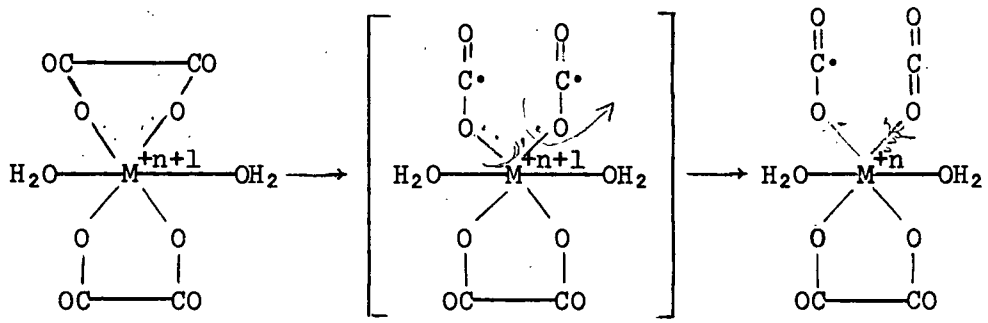


Figure 8. Effects of Pyrophosphate and Oxalate on Peroxides Produced from MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2



Such a mechanism could have been operating between trace metal ions and the added oxalate in the MBG system. The radicals produced may then have initiated glucoside degradation.

This adea about the action of oxalate was supported by the results of Reaction (107)(Fig. 7). When both oxalate and ferric ion were added to the MBG system, a

synergistic accelerative effect was observed. Glucoside degradation proceeded at an increased rate for several hours. The maximum concentration of peroxide intermediate was lower than when no additives were used (Fig. 8), but the peroxide was not completely eliminated, as it was when only ferric ion was added [Reaction (86), Fig. 6]. Oxalate was indeed interacting with ferric ion. In this case, both the ferric-peroxide and the ferric-oxalate interactions could have been providing initiating radicals.

The addition of both ferric and oxalate ions also increased the production of "other peroxides" (Fig. 8), above the level observed when ferric ion alone was added. These additional peroxides were evidently not the direct result of oxalate oxidation, since they were not produced when oxalate alone was added. Instead, they seemed to result from a ferric-oxalate interaction. Perhaps there is a general correlation between generation of radicals (from oxalate or from the peroxide intermediate) and the production of "other peroxides."

The unforeseen accelerative effects of oxalate demonstrated that substances which might be expected to be relatively inert may, under oxygen bleaching conditions, profoundly affect carbohydrate degradation. This emphasizes the importance of other organic components in the bleaching system. Pearl and Beyer (51) found oxalate to be among the major products of alkali lignin autoxidation. The influence of oxidizable organic compounds was studied by Ericsson, et al. (52), who reported that phenolic lignin models increased the metal ion catalysis of cellulose degradation in oxygen-alkali. This was analogous to the synergistic ferric-oxalate effect observed in the present work. Ericsson, et al. (52) and Samuelson and Stolpe (53) also noted that various organic complexing agents sometimes accelerate carbohydrate degradation in oxygen-alkali.

EFFECTS OF ADDED HYDROGEN PEROXIDE

The addition of hydrogen peroxide at the start of a reaction of MBG caused a temporary increase in the initial glucoside degradation rate, as shown in Fig. 9. This would be expected if peroxide decomposition were causing initiation of MBG degradation.

The results of peroxide analyses when H_2O_2 was added at the start of MBG reactions are presented in Fig. 10. When both magnesium and peroxide were present initially [Reaction (137)], the peroxide concentration increased at the same rate as that observed when no H_2O_2 was added (about 0.2 mM/hr.). The similarity in peroxide behavior in Reaction (137), as compared with the other reactions involving magnesium, is apparent from Fig. 10. This shifting of the peroxide intermediate curve to the left is what one would expect if a given amount of the intermediate were present at zero time. The implication is that the peroxide intermediate is hydrogen peroxide. Further evidence for this hypothesis is presented in later sections.

EFFECTS OF GLUCOSIDE CONCENTRATION AND OXYGEN PRESSURE

As mentioned above, the postinduction period MBG degradation rate was essentially unaffected by those additives which influenced the initial rate. That is, the final rate observed with no additives was the same as that found in the presence of magnesium, ferric ion, or hydrogen peroxide (Appendix IV, Table V).

If $[G]_o$ is taken as the glucoside concentration at reaction time t_o after the initial period, in the presence or absence of the above additives, the glucoside data closely fit the relationship

$$1/[G]_o = 1/[G] + q(t-t_o), \qquad (9)$$

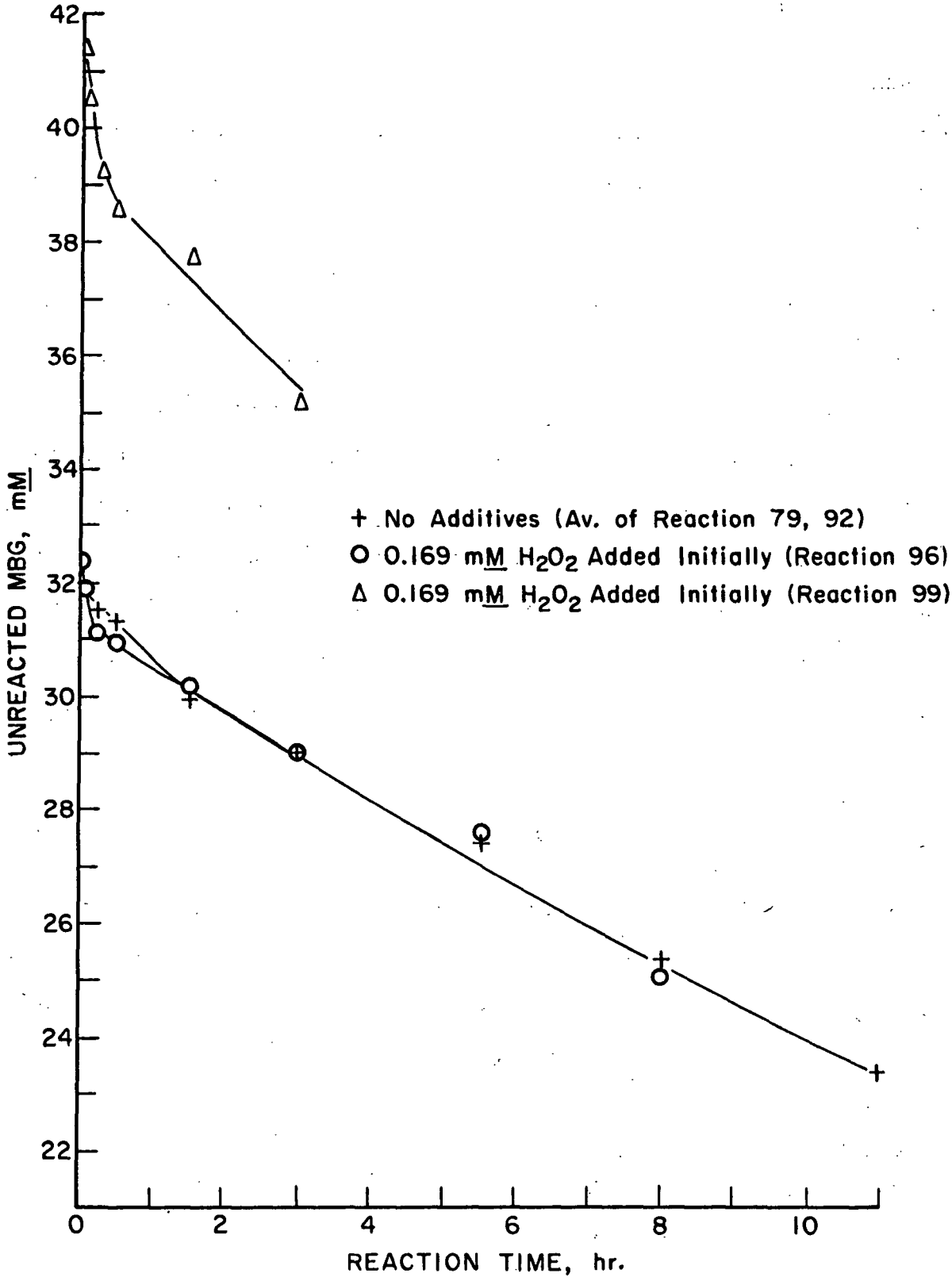


Figure 9. Effect of Added H_2O_2 on the Degradation of MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2

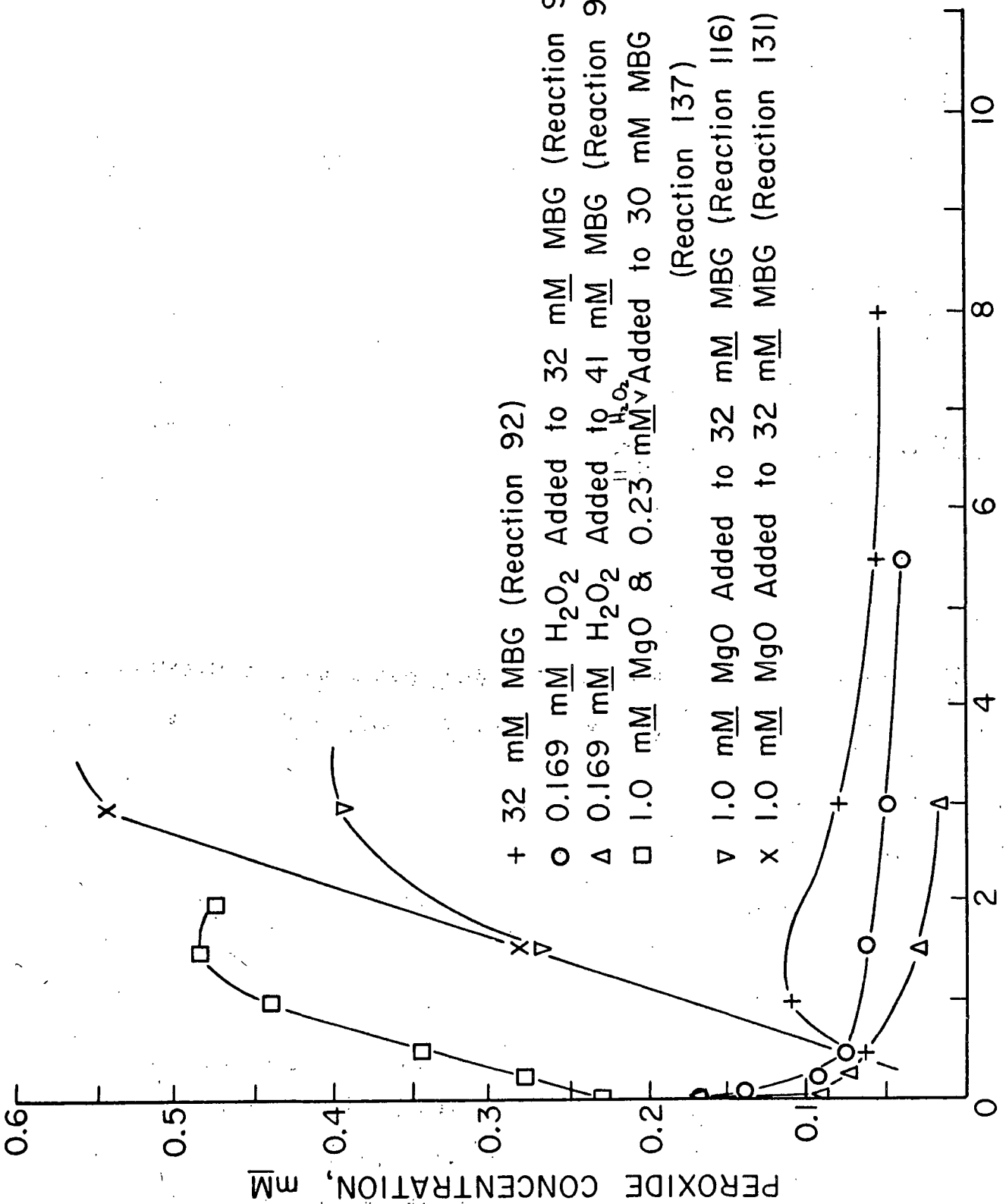


Figure 10. Effect of Added H_2O_2 on the Peroxide Intermediate Produced from MBG in 5% NaOH at 120°C., Under 75 p.s.i. O_2

where [G] is the MBG concentration at reaction time t, and q is a constant. Equation (9) is the relationship for a reaction which is second-order in [G], without autocatalysis or autoinhibition (54).

When postinduction period reaction rates (Appendix IV, Table V) were plotted against [G] values on log-log paper, slopes were generally very near 2. This second-order dependence on glucoside concentration at 20-40 mM was in agreement with McCloskey's findings at 10 mM MBG (11).

Figure 11 depicts the MBG degradation observed at various oxygen pressures. Decreasing the oxygen pressure from 75 to 55 p.s.i. caused a significant decrease in degradation rate. Increasing it to 95 p.s.i., however, produced practically the same rate as that observed at 75 p.s.i. Reaction (134) was run with 50 mM MBG under 95 p.s.i. oxygen. In this case, the rate was only slightly greater than that predicted for 75 p.s.i. at this MBG concentration, using the second-order relationship in glucoside. Thus, the oxygen dependency at these glucoside levels is between zero and first-order. Michie and Neale (29) reported an identical oxygen dependency in alkali cellulose aging. McCloskey (11), on the other hand, reported an approximately first-order dependency on oxygen pressure with 10 mM MBG.

The effects of oxygen pressure and glucoside concentration on the production of the peroxide intermediate were as expected (Fig. 12). Although there is a high degree of uncertainty, the use of lower oxygen pressure generally decreased the rate of peroxide production. The run with highest MBG concentration and oxygen pressure [Reaction (134)] exhibited the highest maximum peroxide level.

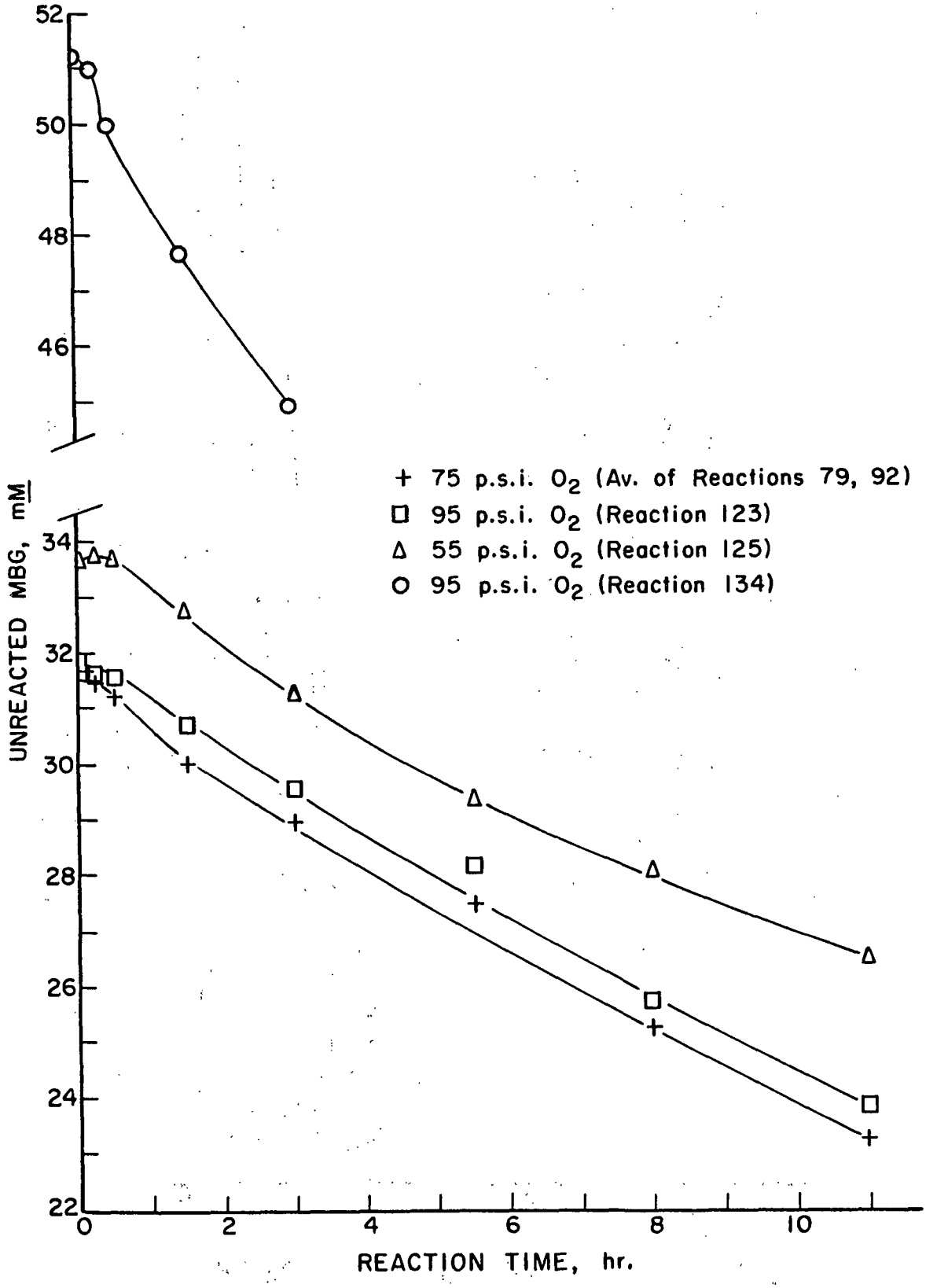


Figure 11. Effect of Oxygen Pressure on the Degradation of MBG in 5% NaOH at 120°C.

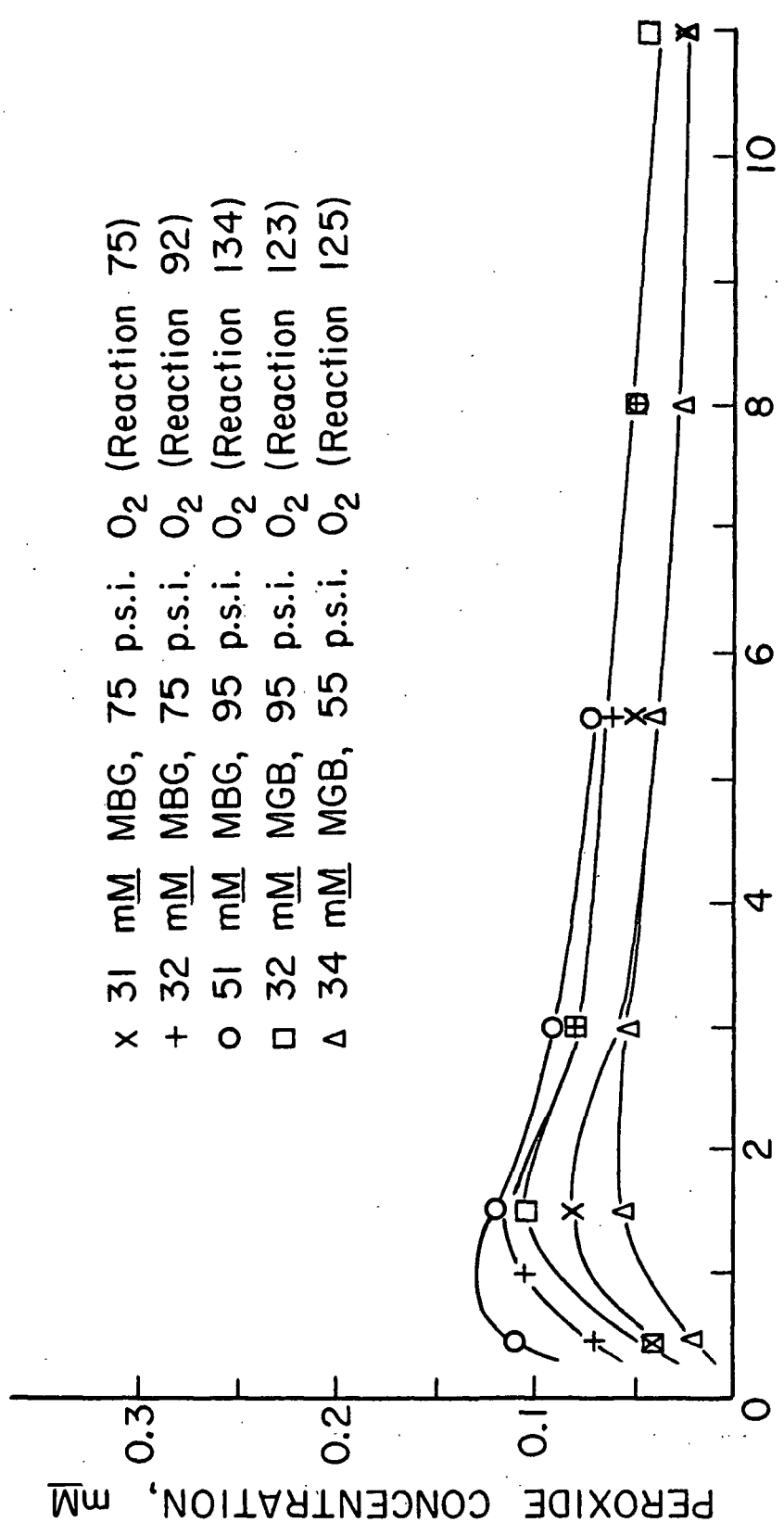


Figure 12. Effect of Oxygen Pressure on the Production of Peroxide Intermediate from MBG in 5% NaOH at 120°C.

STABILITY OF PEROXIDES IN 5% SODIUM HYDROXIDE

PEROXIDE INTERMEDIATE FROM GLUCOSE COMPARED WITH HYDROGEN PEROXIDE AT 35°C.

In order to further test the idea that the peroxide intermediate produced from MBG is hydrogen peroxide, the stabilities of these two peroxides were compared, in the same magnesium sulfate solution, at 35°C. The procedure was outlined in the Experimental section, and analytical results are tabulated in Appendix IV, Table VII. The MBG peroxide intermediate had a first-order decomposition constant of 9.2×10^{-4} hr.$^{-1}$ Hydrogen peroxide, added to the same solution after the original peroxide had decomposed, exhibited a decomposition constant of 8.8×10^{-4} hr.$^{-1}$ under the same conditions. Thus, the "known" and the "unknown" peroxides decomposed at very nearly the same rate.

HYDROGEN PEROXIDE AT 20°C.

The stabilizing effects of various amounts of magnesium sulfate on alkaline hydrogen peroxide at 20°C. were investigated. Analytical results are tabulated in Appendix IV, Table VIII, and presented graphically in the semilog plot of Fig. 13. The first-order decomposition constants k, listed in Table I, were obtained from slopes of Fig. 13. Also listed in Table I are ratios of the decomposition constants obtained in the presence of i mM magnesium (k_i), to the decomposition constant in the absence of magnesium (k_o). These decomposition constant ratios R_i provide a measure of the degree of stabilization effected by the added magnesium. Reference will be made to these R_i values in the discussion of polarographic results.

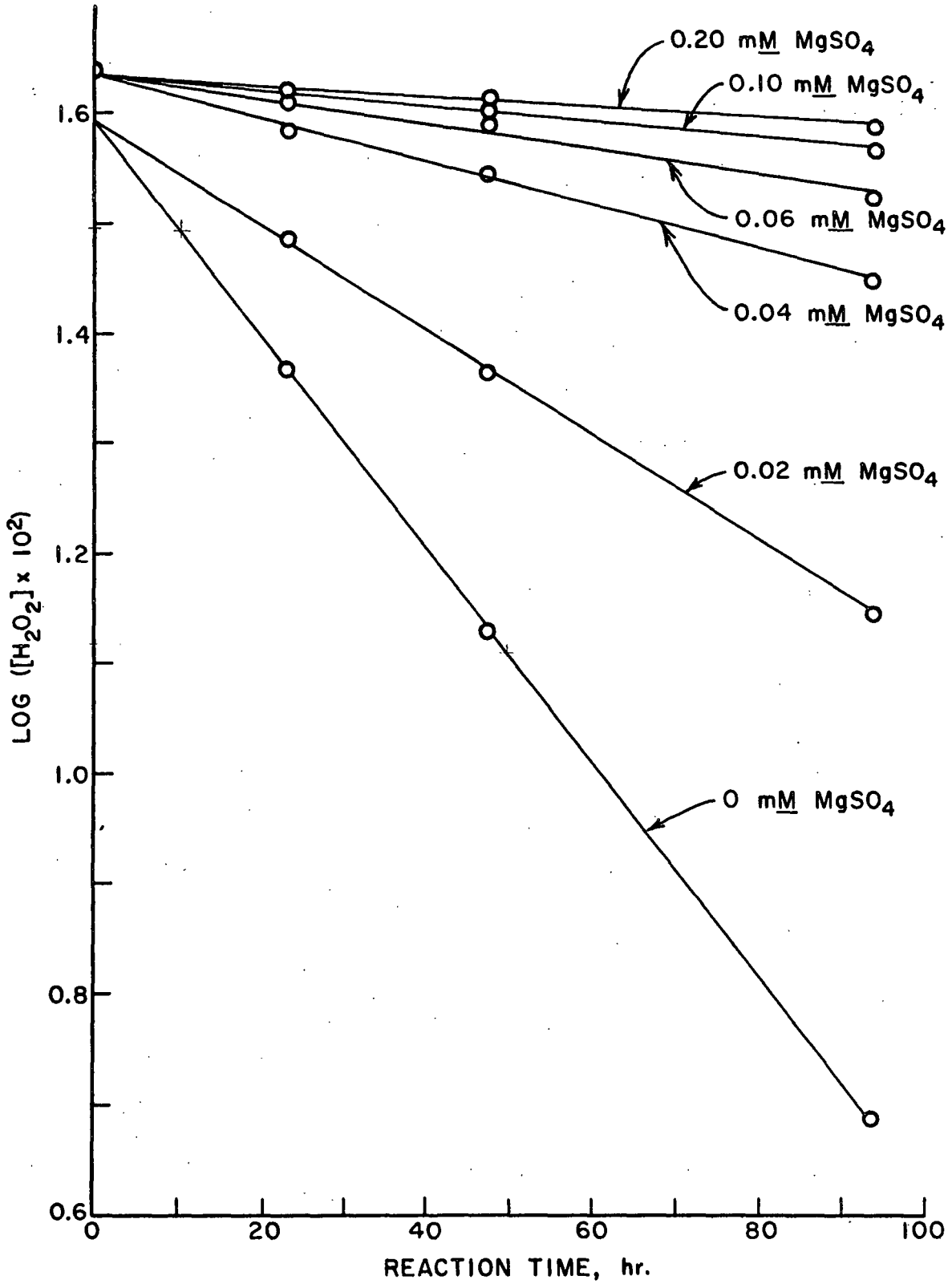


Figure 13. Effect of $MgSO_4$ on the Decomposition of H_2O_2 in 5% NaOH at 20°C.

TABLE I

EFFECT OF $MgSO_4$ ON H_2O_2 DECOMPOSITION CONSTANTS IN 5% NaOH AT ~~120~~ ≈1.25N 20°C.

$[H_2O_2]_o$ = 0.43 mM

i = mM $MgSO_4$	k_i = d log $[H_2O_2]/dt$, $hr.^{-1}$	$R_i = k_i/k_o$
0.00	9.6×10^{-3}	1.000
0.02	4.8×10^{-3}	0.500
0.04	1.9×10^{-3}	0.200
0.06	9.7×10^{-4}	0.100
0.10	6.5×10^{-4}	0.068
0.20	4.2×10^{-4}	0.044

HYDROGEN PEROXIDE AT 120°C.

In connection with Fig. 3, the reader will recall that the maximum concentration attained by the peroxide intermediate varied between runs which were supposedly duplicates. Apparently, the stability of this peroxide was being affected by an uncontrollable factor, both in the presence and absence of added magnesium. If this peroxide is hydrogen peroxide, it too should exhibit a variable stability when added to a similar medium at 120°C. As shown in Fig. 14 and Table IX of Appendix IV, this was found to be the case.

In the presence of magnesium, hydrogen peroxide stability was fairly reproducible when there was no MBG in the solution [Reactions (130), (136b), and (138d). The presence of oxygen had little or no effect. The peroxide decomposition was preceded by an induction period.

When both magnesium and MBG were present, however, H_2O_2 decomposition was more rapid, and much less predictable [Reactions (136a), (136c), (138c), and (139c)].

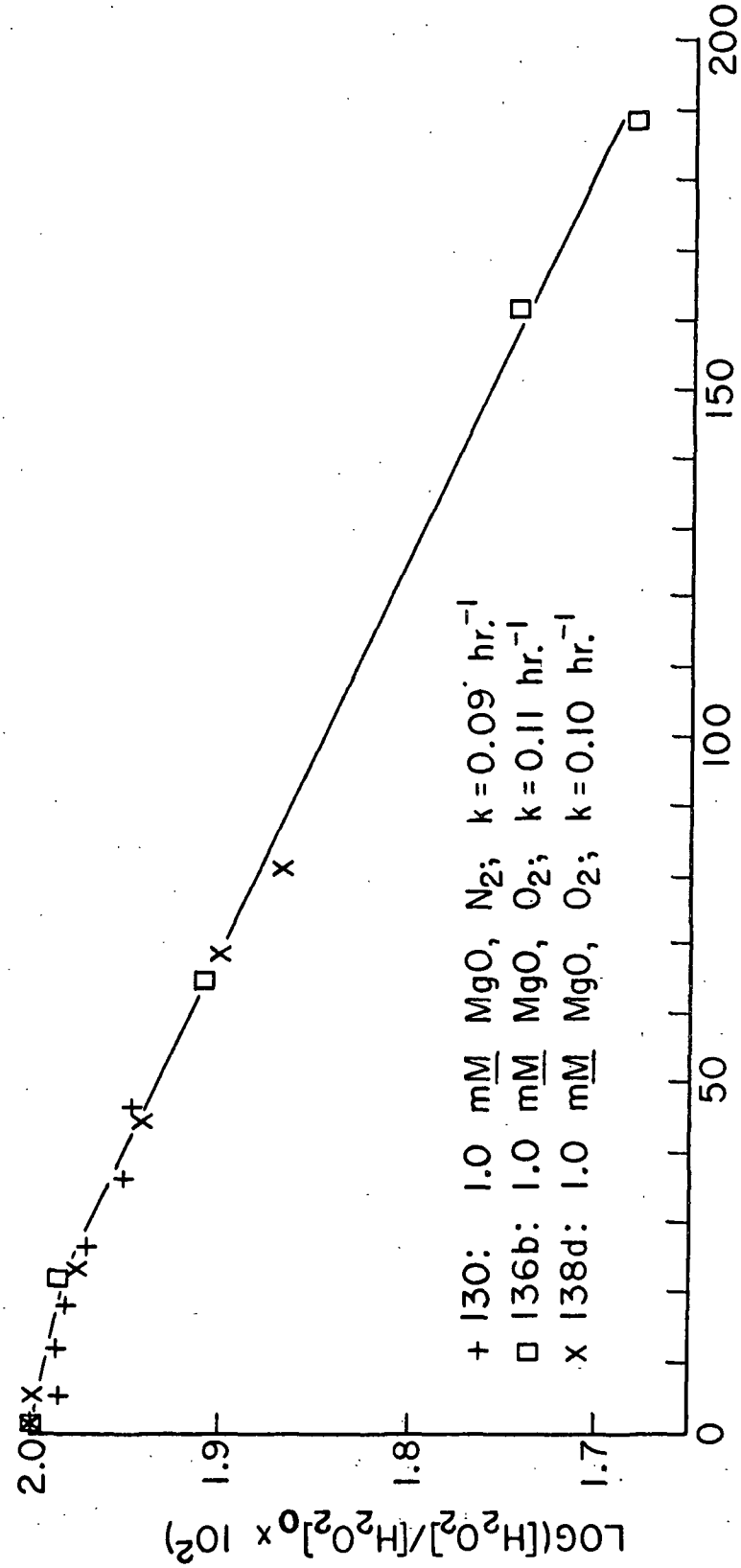


Figure 14a. Decomposition of H_2O_2 in 5% NaOH at 120°C.

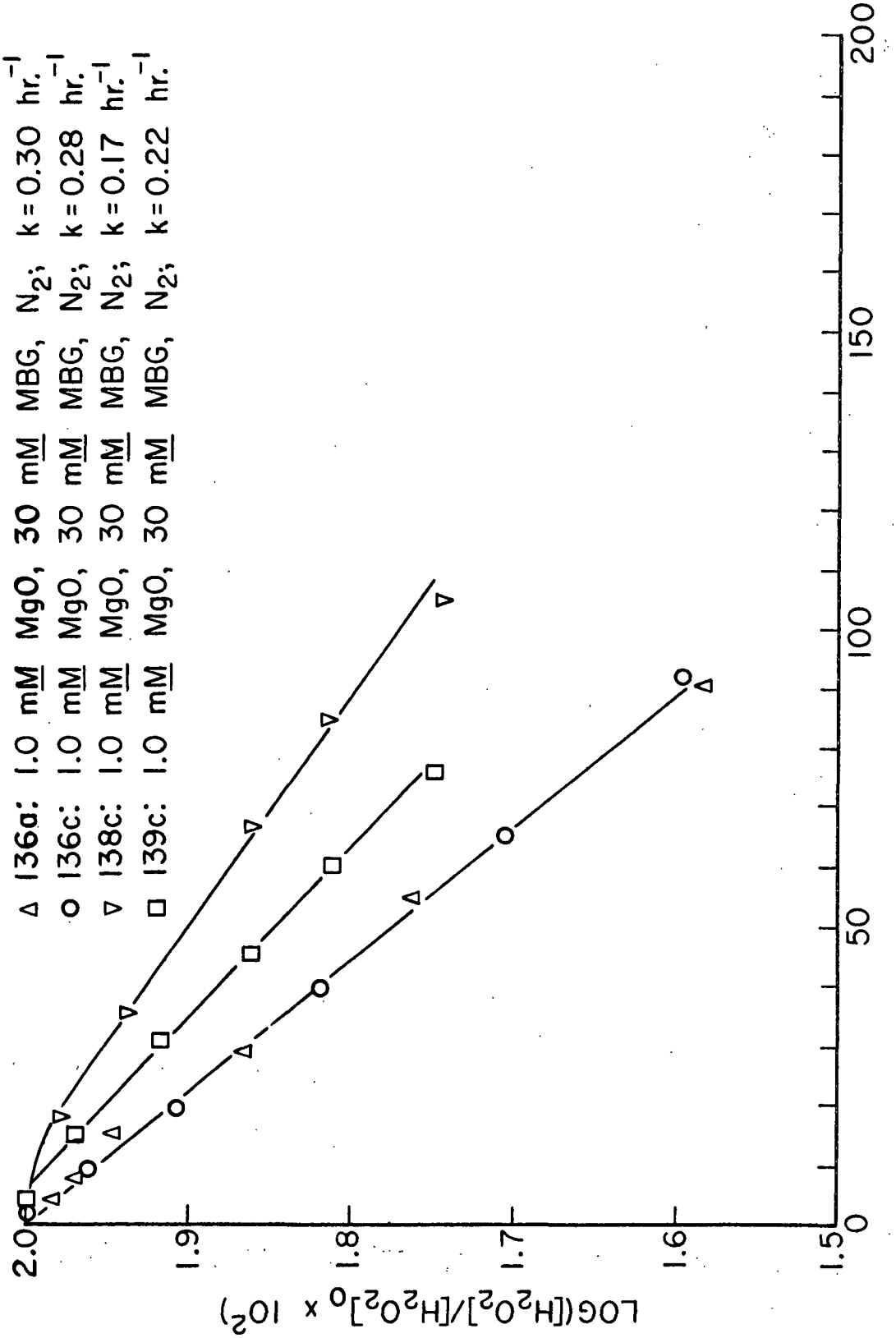


Figure 14b. Decomposition of H_2O_2 in 5% NaOH at 120°C.

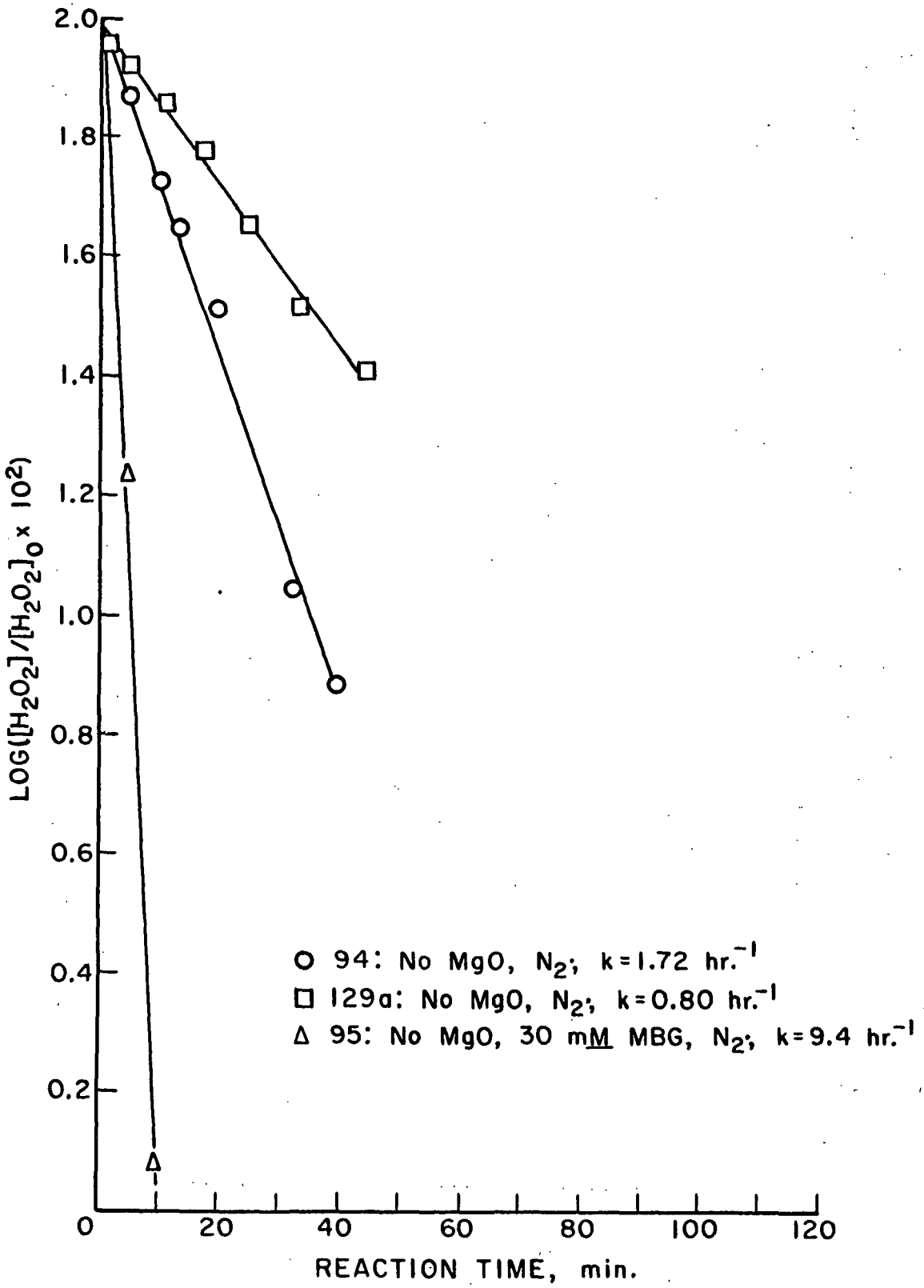


Figure 14c. Decomposition of H_2O_2 in 5% NaOH at 120°C.

The induction period varied from 0 to ca. 20 minutes, and the decomposition constants varied from 0.17 to 0.30 $hr.^{-1}$

Peroxide decomposition was, of course, much more rapid in the absence of magnesium [Reactions (94), (95), and (129a)]. Here again, reproducibility was very poor, and the presence of MBG brought about faster peroxide decomposition.

The reason for the accelerating influence of MBG on H_2O_2 decomposition is uncertain. Perhaps the glucoside acts as a chain transfer agent in a radical decomposition reaction.

The variability in H_2O_2 decomposition rates is probably connected with the kinds and amounts of unavoidable trace contaminants in the solutions. Whatever the cause of this phenomenon, the significant point is that it is characteristic of both the peroxide intermediate and hydrogen peroxide itself.

PEROXIDE PRODUCED FROM D-GLUCOSE IN OXYGEN-ALKALI AT ROOM TEMPERATURE

Glucose was used as a model to study the effects of magnesium on the reaction of a reducing sugar in oxygen-alkali. Since reducing sugars are degraded much more rapidly than nonreducing sugars, a lower temperature was employed (22 ± 2°C.). The reactions were followed by measuring peroxide production (Fig. 15 and Table VI of Appendix IV). This peroxide gave the same type of immediate color response as that exhibited by H_2O_2 and by the MBG peroxide intermediate. None of the glucose test solutions increased in absorbance with time, to indicate the presence of any other peroxides.

Magnesium had no significant effect on the initial rate of peroxide production. In the absence of other peroxide stabilizers (Curves E and F), magnesium exhibited a slight stabilizing effect. When pyrophosphate was present however (Curves A and

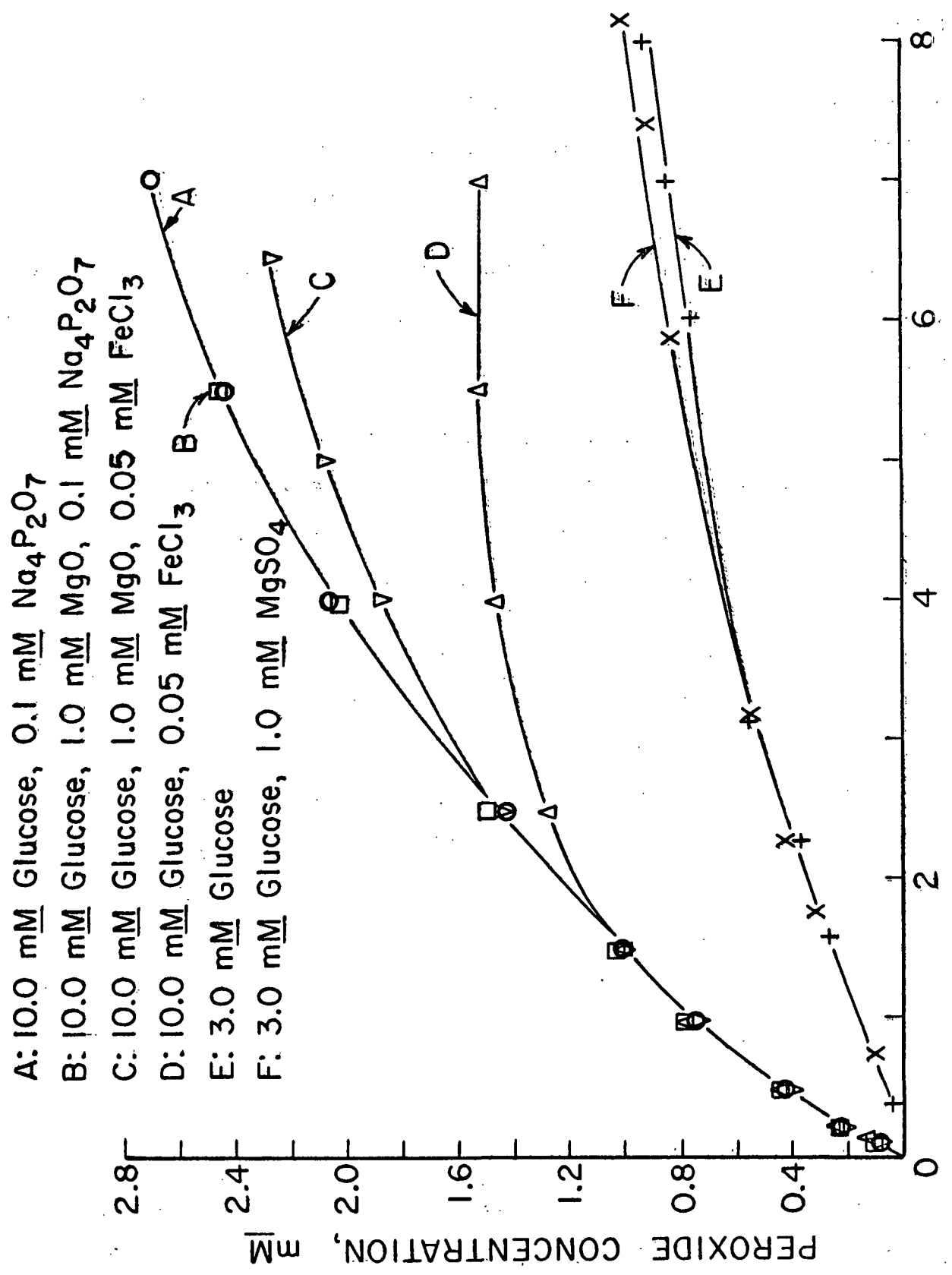


Figure 15. Peroxide Produced from D-Glucose in 5% NaOH at 22°C., Under 75 p.s.i. O_2

B), magnesium had no additional influence. The addition of 0.05 mM ferric chloride did not affect initial peroxide production, but it decreased its stability. Magnesium tended to counteract the effects of added ferric ion.

Bamford and Collins (36) noted that this room temperature glucose reaction was unaffected by most catalytic ions, and appeared to be an ionic reaction. The present observation, that it is also unaffected by magnesium, is consistent with this. These workers (36) also reported that the reaction is first-order in glucose, and that the initial rate of peroxide production equals the concurrent rate of glucose degradation. The curvature of lines A, B, E, and F of Fig. 15, however, indicate a net rate of peroxide production which is less than first-order in glucose. This either means that the peroxide undergoes a subsequent reaction which is unaffected by magnesium, or some glucose molecules react with oxygen without producing the stable peroxide. The latter possibility is more consistent with results reported by other workers (36,37). The implications of this will be discussed further in a later section.

POLAROGRAPHY

APPLICABILITY OF POLAROGRAPHY

No attempt will be made here to consider fundamentals of polarographic theory, instrumentation, or technique. The reader who is unfamiliar with these topics is referred to such works as those of Meites (55), Zuman (56), and Kolthoff and Lingane (57).

Polarography can often provide a value for the diffusion coefficient of a substance, and hence a relative measure of the size of a reducible species in an electrolyte solution. Moreover, the half-wave potential at which any substance

is reduced at the dropping mercury electrode (D.M.E.) is a measure of its ease of reduction. Hence, if one could obtain, in the same electrolyte, polarograms of H_2O_2 and the peroxides produced from MBG and glucose, they could be compared in terms of oxidizing power and diffusion coefficients. The latter parameter in particular would be expected to differ between H_2O_2 and any (higher molecular weight) organic peroxide.

Polarography also provides a means for testing the theory of formation of a magnesium-peroxide complex. When a reducible substance is complexed, its half-wave potential generally acquires a more negative value (it becomes harder to reduce); and its diffusion coefficient decreases if the larger complexed form is the species being reduced (57).

Polarography was found to be directly applicable to MBG reaction samples. The 1.25N sodium hydroxide solutions were excellent electrolytes (55). Neither the starting material (a nonreducing sugar) nor the reaction products (hydroxy acids) were reduced in the 0 to -2 volt range (57). Thus, it was possible to obtain a polarogram of only peroxides, without interference from other substances in solution. The polarographic study of glucose peroxides also proved to be feasible. Glucose does give a small polarographic response (57), but interference from this reducing sugar could be eliminated by using small sugar concentrations.

EFFECTS OF EXPERIMENTAL PARAMETERS ON HYDROGEN PEROXIDE WAVES

Figure 16 is a representation of a polarogram of hydrogen peroxide in 1.25N sodium hydroxide solution. The final current rise is caused by reduction of sodium ion (57). The curves depict the top of the oscillations on the recorder chart, i_{max}, or the maximum current attained just before the mercury drops fall.

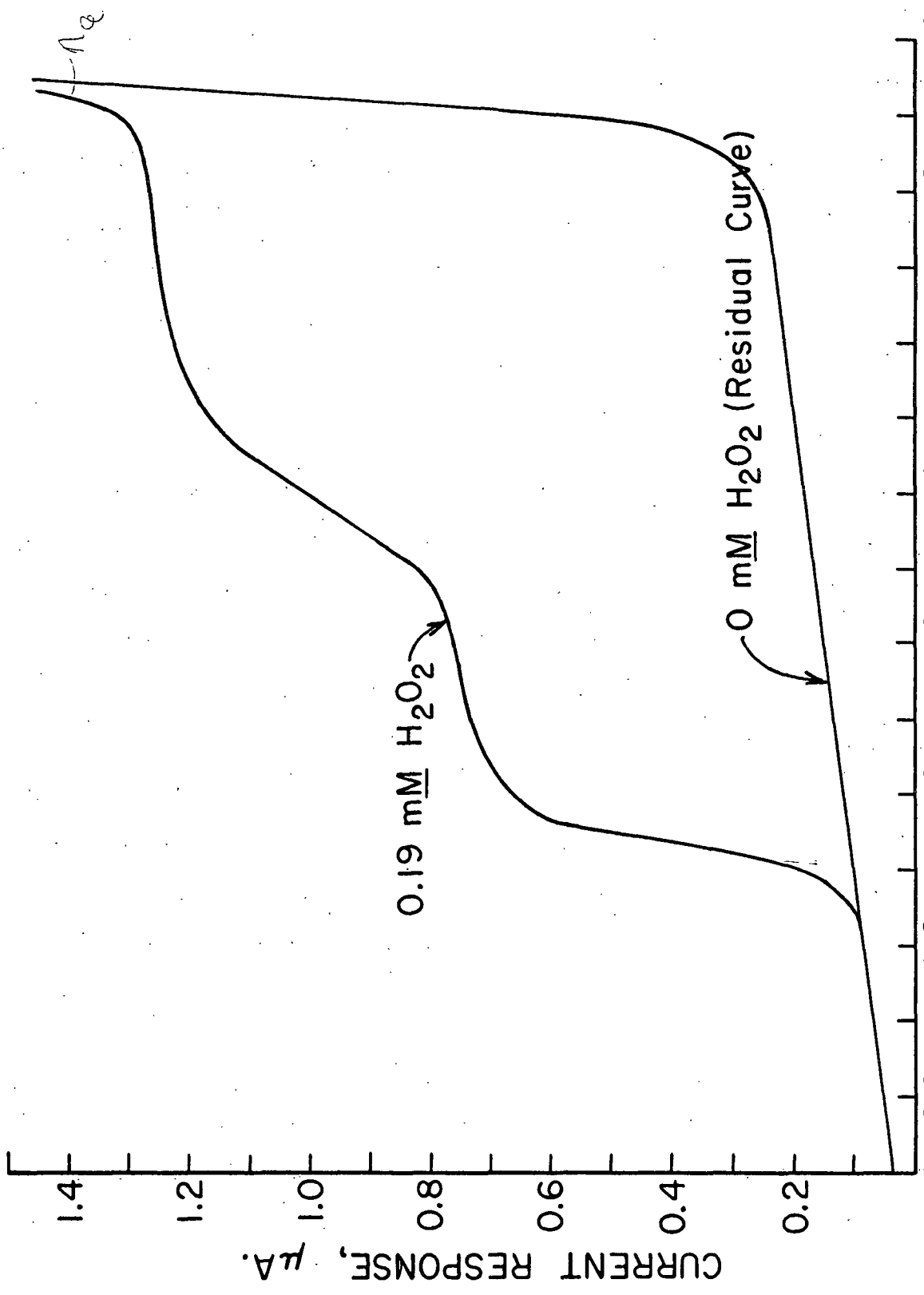


Figure 16. Polarogram of H_2O_2 in 5% NaOH as Supporting Electrolyte

Although a more common practice is to measure and report the average of the oscillations $\bar{i}$, this method was chosen for its simplicity, and because the instrument manual[2] recommends it. Generally, $\bar{i} = 6i_{-max}/7$ (57).

Hydrogen peroxide produced two waves in 1.25N sodium hydroxide: one at ca. -0.95 v. vs. S.C.E., and another at ca. -1.4 v. The current response i_{-max} of the first wave was taken as the limiting current minus the residual current at -1.1 v. The height of the second wave was likewise measured at -1.8 v. Table X of Appendix IV lists values of i_{-max} at these two potentials, for all polarograms discussed herein.

As seen in Fig. 17, the current response at -1.8 v. increased linearly with peroxide concentration. The wave height measured at -1.1 v., however, approached a limit as H_2O_2 concentration increased. According to Zuman (56), a linear response to concentration is typical of diffusion currents, and most kinetic currents. (With a diffusion current, the response is controlled by the rate of diffusion of the reducible species to the mercury electrode; a kinetic current is controlled by the rate of a chemical reaction at the electrode.) On the other hand, catalytic currents (kinetic currents which are controlled by a catalyzed chemical reaction), and adsorption currents (controlled by adsorption phenomena at the electrode), reach limiting values with increasing concentration. Thus, the first H_2O_2 wave is either a catalytic or an adsorption wave, while the second wave may be either diffusion or kinetic controlled.

Further information about the current response was obtained by varying the height of the mercury column above the D.M.E. Diffusion currents are proportional to the square root of mercury column height, after correcting for mercury

[2] Instruction Manual S-29310, E. H. Sargent & Co., 4647 W. Foster, Chicago, Ill.

back pressure according to Equation (10):

$$h_{corr} = h - 3.1/m^{1/3}\ t^{1/3}\ \text{cm.}, \qquad (10)$$

where $\underline{m}$ and $\underline{t}$ are mercury flow rate and drop time, respectively. Kinetic currents are independent of $\underline{h}_{corr}$, and adsorption currents are proportional to $\underline{h}_{corr}$ (not to its root). Catalytic currents exhibit various dependencies on $\underline{h}$ (56). In the present work, it was found that both H_2O_2 wave heights were proportional to the square root of corrected mercury column height. Therefore, the first wave represents a catalytic current, and the second a diffusion current.

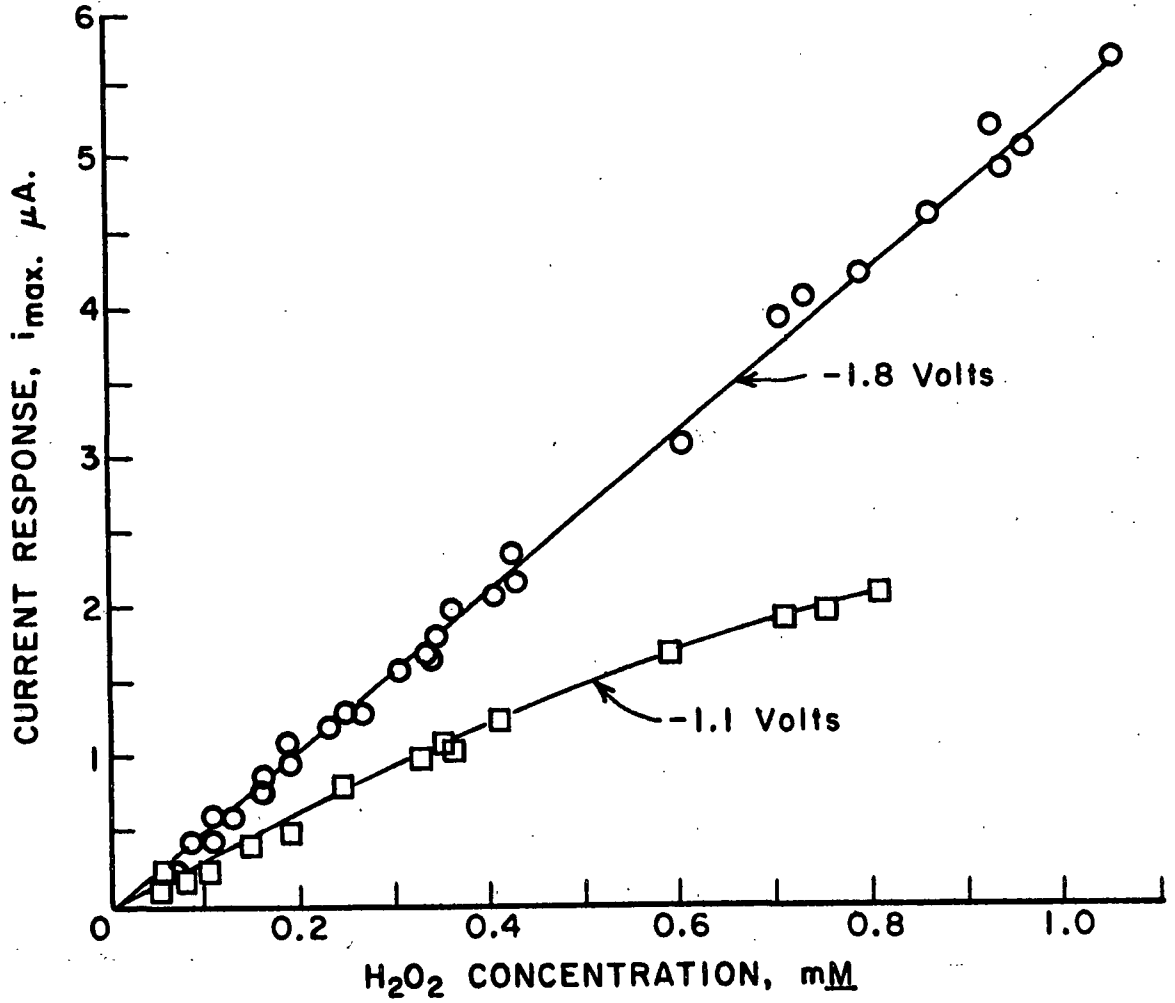


Figure 17. Polarographic Response of H_2O_2 in 5% NaOH

Many kinetic currents show a large temperature dependency (up to 30% increase per °C.). Most other currents exhibit temperature coefficients of less than 2%/°C. (55-57). The temperature coefficients of both hydrogen peroxide waves here was found to be +1-2%/°C.

Thus, the total current response was controlled by the rate of diffusion of peroxide in the electrolyte solution. Hence, the diffusion coefficient D of the peroxide could be estimated from the Ilkovic equation, $I = 607nD^{\frac{1}{2}}$, where n is the number of Faradays of electricity per mole, and I is the diffusion current constant (57). This latter parameter is defined by

$$I = i_d/Cm^{2/3}t^{1/6}, \qquad (11)$$

where i_d is the average diffusion current in μA. during the life of a mercury drop, C is the concentration of the reducible substance in mM, m is the mercury flow rate in mg./sec., and the t is the drop time in sec. Generally, I is constant for a given substance in a given electrolyte (57). For hydrogen peroxide reduction, n = 2 (57). The value here of $i_d/C = 6(i_{max,\ -1.8})/7C$ is 4.68, from the slope of Fig. 17. Substitution of this and other experimental values into Equation (10) and the Ilkovic equation yields a value for I = 3.0, and $D = 6.1 \times 10^{-6}$ cm.^{2}/sec.

EFFECTS OF ADDITIVES ON HYDROGEN PEROXIDE WAVES

Figure 18 qualitatively illustrates the effects of various additives on a polarogram of H_2O_2 in 1.25N sodium hydroxide. Quantitative data are presented in Table X of Appendix IV. The addition of sodium tartrate diminished the first wave, but magnesium sulfate essentially eliminated this catalytic response. The presence of ferric ion, on the other hand, caused the catalytic wave to predominate.

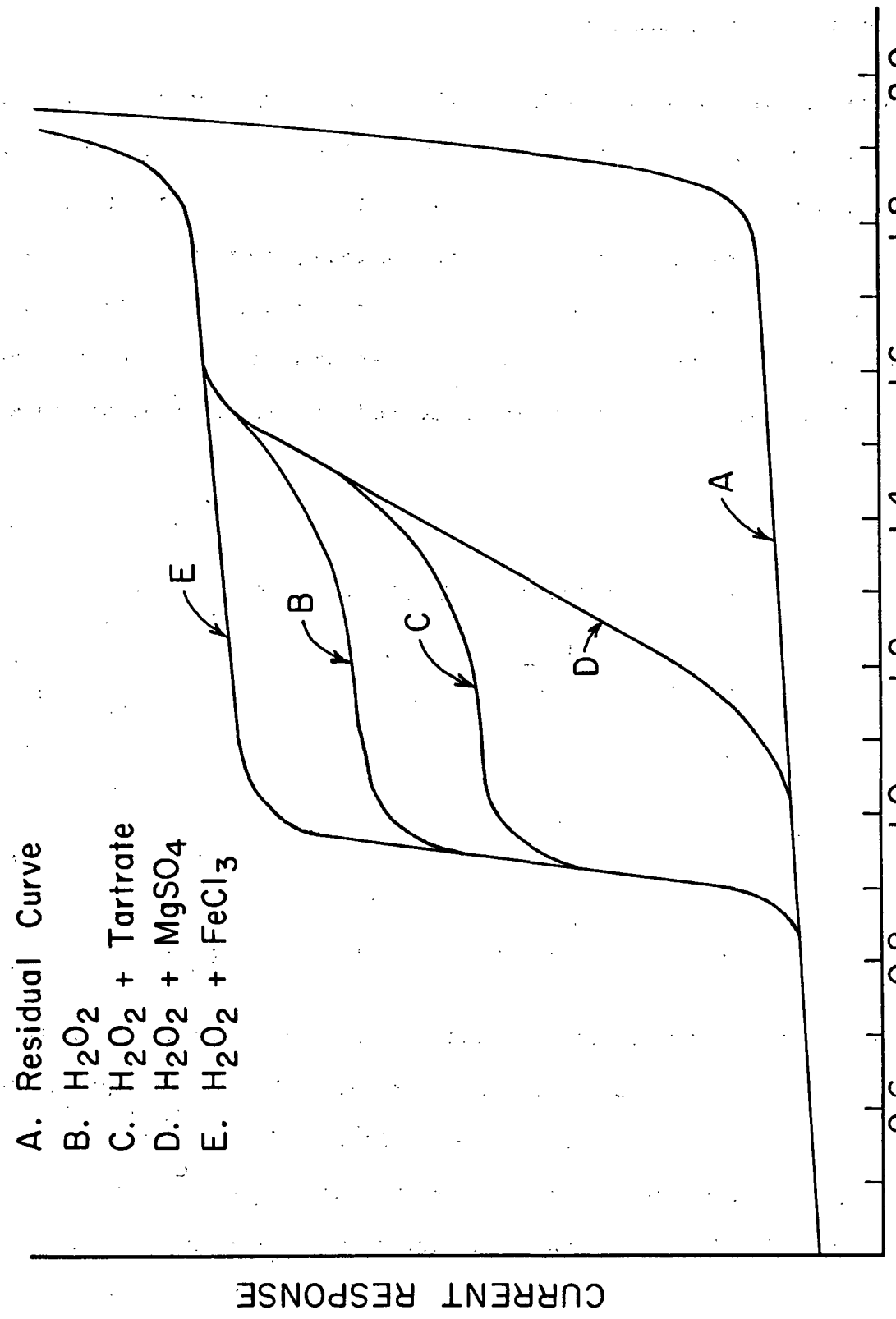


Figure 18. Typical Polarograms of H_2O_2 in 5% NaOH Containing Various Additives

The polarogram of H_2O_2 was not affected by the presence of MBG (30 m$\underline{M}$), glucose (10 m$\underline{M}$), or sodium pyrophosphate (0.1 m$\underline{M}$).

Data are presented in Tables II and III, to illustrate quantitatively the effects of various amounts of tartrate and magnesium on polarograms of H_2O_2. The value of ρ_j, the ratio of the current response at -1.1 v. to that at -1.8 v., gives a numerical indication of the importance of the catalytic wave. R_j is the ratio of ρ_j, at a tartrate or magnesium concentration of j m$\underline{M}$, to the value of ρ in the absence of tartrate or magnesium (ρ_o). R_j serves as a reciprocal measure of the relative effectiveness of the tartrate or magnesium in diminishing the catalytic wave.

TABLE II

EFFECTS OF TARTRATE ON RELATIVE H_2O_2 WAVE HEIGHTS

Electrolyte	j = m$\underline{M}$ Tartrate	i_{max} at -1.1 v.	i_{max} at -1.8 v.	$\rho_j = i_{-1.1}/i_{-1.8}$	$R_j = \rho_j/\rho_o$
Ca. 0.25 m$\underline{M}$ H_2O_2; 5% NaOH	0.0	0.975	1.456	0.670	1.000
	1.0	0.883	1.416	0.624	0.931
	4.0	0.710	1.382	0.513	0.756
	10.0	0.541	1.353	0.400	0.598
	23.8	0.320	1.286	0.249	0.372

Magnesium sulfate was about 100 times as effective as sodium tartrate in lowering the height of the catalytic wave. The effectiveness of magnesium was inhibited, however, when tartrate was present in the same solution. This may be due to the formation of the fairly stable magnesium-tartrate complex (50). Magnesium did have the ability to counteract the effects which ferric ion had on the catalytic wave.

TABLE III

EFFECT OF $MgSO_4$ ON RELATIVE H_2O_2 WAVE HEIGHTS

Electrolyte	j = mM $MgSO_4$	i_{max} at -1.1 v.	i_{max} at -1.8 v.	$\rho_j = i_{-1.1}/i_{-1.8}$	$R_j = \rho_j/\rho_o$
Ca. 0.05 mM H_2O_2; 5% NaOH	0.000	0.264	0.292	0.905	1.000
	0.013	0.231	0.276	0.838	0.926
	0.038	0.076	0.211	0.360	0.398
	0.050	0.036	0.200	0.180	0.199
	0.062	0.018	0.200	0.090	0.100
Ca. 0.70 mM H_2O_2; 5% NaOH	0.000	2.001	4.032	0.496	1.000
	0.012	1.410	3.936	0.358	0.712
	0.035	0.513	3.864	0.133	0.268
	0.047	0.396	3.819	0.104	0.210
	0.058	0.309	3.780	0.082	0.160
Ca. 0.23 mM H_2O_2; 8 mM tartrate; 5% NaOH	0.000	0.580	1.226	0.473	1.000
	0.012	0.490	1.193	0.410	0.867
	0.037	0.349	1.156	0.302	0.640
	0.049	0.259	1.145	0.226	0.479
	0.061	0.192	1.127	0.170	0.360
Ca. 2.4 mM H_2O_2; 0.01 mM $FeCl_3$; 5% NaOH	0.000	13.24	14.96	0.887	1.000
	0.063	7.40	12.99	0.570	0.642
	0.125	1.86	12.49	0.149	0.168
	0.189	1.05	12.01	0.087	0.099

When a sodium hydroxide solution was not nitrogen-purged, a polarogram of the dissolved oxygen could be obtained. In this electrolyte, oxygen gave three waves. The first exhibited a half-wave potential of ca. -0.1 v. The second and third waves showed the same relative heights and half-wave potentials as those of H_2O_2 in this electrolyte. The sum of the heights of the second and third oxygen waves was equal to that of the first wave.

In nonalkaline electrolytes, oxygen is normally reduced to give two waves of equal height (57). The first wave represents a reduction of oxygen to hydrogen peroxide, and the second a reduction of the peroxide to hydroxyl ions. Other workers (57) have noted the creation of a third oxygen wave, similar to that encountered here, when certain iron compounds were present in neutral or

acidic electrolytes. Thus, the addition of substances like hemoglobin, heme, etc., did not affect the first oxygen wave, but caused part of the last wave (H_2O_2 reduction) to shift to more positive potentials. This "prewave" of hydrogen peroxide was attributed to a catalytic effect, whereby the iron caused some of the peroxide to be more easily reduced. The height of the catalytic wave was proportional to the concentrations of H_2O_2 and the catalyst.

Hence, the first hydrogen peroxide wave observed in the present study was apparently caused by the catalytic action of trace metal ions in the strong alkali. This is supported by the effects of added ferric ion and of tartrate. The second hydrogen peroxide wave represents the normal uncatalyzed reduction of H_2O_2 to hydroxyl ions. Magnesium did not affect either the half-wave potential or the magnitude of this second wave. This implies that there is no direct magnesium-peroxide interaction. If magnesium were inhibiting H_2O_2 decomposition by forming a more stable peroxide or complex, the normal H_2O_2 reduction wave would be shifted to a more negative potential. That is, the stabilized peroxide would be more difficult to reduce. Moreover, the larger size of a complexed peroxide would cause a decrease in the diffusion coefficient D, and in the diffusion current constant I. As mentioned in connection with Fig. 17, however, the diffusion current constant for H_2O_2 was not affected by magnesium, or any other additive.

Thus, it appears that magnesium compounds inhibit alkaline H_2O_2 catalysis by deactivating catalytic ions in the system. When a relatively large amount of ferric chloride (ca. 1 mM) was added to a sodium hydroxide electrolyte, it exhibited a small wave at -0.95 v., and another at -1.55 v. The addition of excess magnesium sulfate to this solution, however, caused both of these iron waves to disappear, indicating that the magnesium-metal ion interaction theory is valid.

Furthermore, as shown in Fig. 19, there is a connection between the ability of magnesium sulfate to diminish the catalytic H_2O_2 wave (R_j of Table III), and its effectiveness in stabilizing the peroxide (R_i of Table I). Thus, the polarographic results are indeed related to peroxide stabilization.

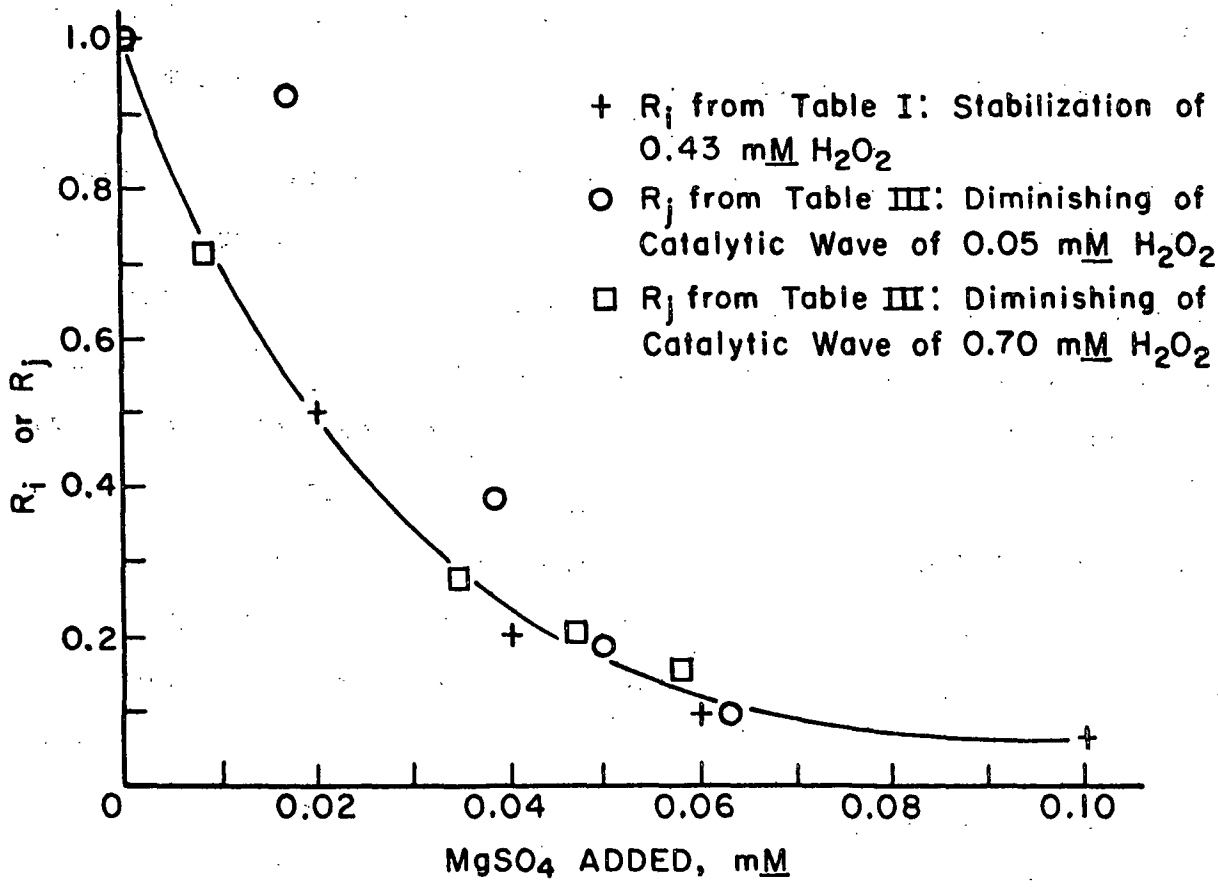


Figure 19. Correlation Between Effect of Magnesium on Catalytic H_2O_2 Wave (R_j) and Stabilization of H_2O_2 (R_i), in 5% NaOH at 20°C.

Concurrent with the present study, other workers also obtained evidence supporting the hypothesis that magnesium stabilizes H_2O_2 by interacting with catalytic ions, rather than with the peroxide itself. Ericsson, et al. (52) noted that 1.25 mM magnesium sulfate effectively inhibited MBG degradation and peroxide decomposition in 65 mM H_2O_2. Because of the concentration difference, a complex between magnesium ion and the peroxide seemed highly unlikely in this alkaline system. Gilbert, et al.

(58) moreover, reported evidence that magnesium ion forms complexes with metal ions via hydroxo- or oxo-bridges.

POLAROGRAPHY OF PEROXIDE PRODUCED FROM METHYL β-GLUCOSIDE

Evidence cited previously indicated that the MBG peroxide intermediate may be hydrogen peroxide. The polarographic data discussed here confirm this theory.

Polarograms were obtained of samples taken after 0, 1.3, and 4.5 hr. reaction of 30 mM MBG in 1.25N sodium hydroxide at 120°C., under 75 p.s.i. oxygen. Data are presented in Table X of Appendix IV. In all cases, the reaction solutions gave the same type of double-wave polarogram as that obtained from hydrogen peroxide in 1.25N alkali (Curve B, Fig. 18). The half-wave potentials of both waves were the same as those of H_2O_2 waves. Concentrations of peroxide intermediate, determined colorimetrically, corresponded to the observed current responses at -1.1 and -1.8 volts (Fig. 17). When a known amount of authentic hydrogen peroxide was added to a reaction solution, the polarographic wave heights increased to levels which corresponded to the total peroxide concentration, according to Fig. 17. The addition of the "known" peroxide to the "unknown" created no new inflection points on the polarogram.

A second MBG reaction, like the first but with 1.0 mM magnesium sulfate present, was carried out. Polarograms of 1.1 and 3.5-hr. samples from this reaction consisted of single waves, identical to those exhibited by alkaline H_2O_2 in the presence of magnesium (Curve D, Fig. 18). Again, the half-wave potential and the diffusion current constant of the peroxide intermediate were the same as those of H_2O_2. Moreover, the addition of H_2O_2 to a reaction solution resulted in the same wave shape, with a larger current response to correspond to the total peroxide concentration.

It is very unlikely that two different peroxides would exhibit the same half-wave potentials for both a normal reduction wave and a catalytic wave. It is even less likely that the diffusion current constants of hydrogen peroxide and an organic peroxide could not be distinguished. To demonstrate this latter point, several polarograms of t-butyl hydroperoxide (TBHP) were obtained in 1.25N sodium hydroxide, containing 0.005% gelatin as a maximum suppressor (57). This organic peroxide gave a single drawn-out wave, from -0.4 to -1.8 v., with a half-wave potential of -0.92 v. The diffusion coefficient of TBHP in this medium was $D = 2.9 \times 10^{-6}$ cm.^{2}/sec., compared with a value of 6.1×10^{-6} cm.^{2}/sec. for H_2O_2. Since a carbohydrate peroxide would probably be of even greater molecular size than TBHP, one would expect that such a peroxide would have an even lower diffusion coefficient. Thus, the fact that the MBG peroxide intermediate and H_2O_2 give identical waves, with indistinguishable values of I and D, leaves little doubt that the intermediate is indeed hydrogen peroxide.

Although the titanium analysis confirmed that the usual "other peroxides" were present in the reaction samples discussed here, they were not detected polarographically. This is not surprising, since they are less reactive than the hydrogen peroxide intermediate. (They do not react readily with iodide ion, and they are only slowly hydrolyzed in strong acid.) Thus, one would expect them to be reduced at more negative potentials, beyond -2 volts.

POLAROGAPHY OF PEROXIDE PRODUCED FROM D-GLUCOSE

Polarograms of D-glucose reaction solutions were obtained, in an attempt to identify the peroxide produced by oxidation of this reducing sugar. Two reactions of 3.0 mM glucose were run at 22°C., in 1.25N sodium hydroxide under 75 p.s.i. oxygen. One of the two reaction solutions contained 1.0 mM magnesium sulfate.

Polarograms of the solutions were obtained after 2.3 and 7.0-hr. reaction in the absence of magnesium, and after 2.3 and 7.4 hr. in the presence of magnesium. Results may be found in Table X of Appendix IV. In all cases, the wave shapes and diffusion current constants of H_2O_2, and of the peroxide produced from glucose, were identical. Since the titanium analysis indicated that no other peroxides were produced from glucose, it appears that the only stable peroxide produced from this reducing sugar is hydrogen peroxide.

SIGNIFICANCE OF RESULTS

THE FUNCTION OF MAGNESIUM COMPOUNDS IN CARBOHYDRATE STABILIZATION

Interaction with Carbohydrate

When McCloskey (11) found that 10 mM MBG was stabilized only slightly by magnesium, it seemed feasible that magnesium could be interacting primarily with a reducing group. Hence, it would be effective only in the presence of reducing sugars. The current research, however, has shown a definite stabilizing effect by magnesium when the MBG concentration is higher. Hence, an interaction between magnesium and a reducing function is clearly an inadequate explanation.

Magnesium ion could theoretically interact with hydroxyl groups away from any reducing function of a carbohydrate, to prevent oxygen attack here. Such an interaction would inhibit peroxide production. The present results, however, both with MBG and with glucose, indicated that magnesium has no effect on peroxide production; it influences only its ultimate degradation. Furthermore, 1.0 mM magnesium was just as effective as 25 mM in stabilizing 30 mM MBG. Thus, any conjectured magnesium-carbohydrate interaction is of little consequence to the degradation.

Free Radical Scavenger

If the reaction involved free radical intermediates, magnesium could inhibit it by acting as a radical scavenger. Such an effect would, however, be expected to decrease the postinduction period rate of degradation. This was not observed in this work with MBG. Ericsson, et al. (52) moreover, reported that magnesium sulfate had no effect on the degradation of MBG by hydroxyl and perhydroxyl radicals. There is, apparently, no evidence to support the idea that magnesium compounds act as radical scavengers.

Interaction with Peroxide Intermediate

There can be little doubt that the MBG peroxide intermediate is hydrogen peroxide. This is supported by its stability in alkali compared with H_2O_2, its behavior in solutions of titanium sulfate and dilute acid, and most of all by polarographic data.

A correlation has been noted, in present data and that of Minor and Sanyer (48), between the start of peroxide depletion and the end of the induction period. The initial carbohydrate degradation was accelerated by the addition of H_2O_2, or species which are readily oxidized to H_2O_2, such as glucose (11,13) or hydroquinone (11). When H_2O_2 was destroyed by iodide ion, initiation was postponed indefinitely. These facts are all consistent with the idea that the more rapid degradation of a sugar such as MBG is initiated as H_2O_2 reacts with some other component of the system. Thus, magnesium could delay initiation by inhibiting this peroxide decomposition reaction. This might occur as a result of magnesium interacting with H_2O_2, or with the other component. It is well known that magnesium inhibits alkaline hydrogen peroxide decomposition in other systems (13,38,39), but the mechanism of this stabilization has not been heretofore established.

The present work provides evidence against the existence of a magnesium-peroxide interaction in this system. In a MBG reaction solution containing magnesium, the insoluble magnesium peroxide MgO_2 could not be detected analytically. Furthermore, the polarographic reduction wave of H_2O_2 retained the same half-wave potential and diffusion current constant when magnesium was added. The polarographic effects of magnesium were instead confined to diminishing a catalytic H_2O_2 wave. In addition, the finding of Ericsson, et al. (52), that the H_2O_2 oxidation of MBG could be retarded by an amount of magnesium which was much less than the amount of peroxide, also refutes the magnesium-peroxide interaction theory.

Interaction with Catalytic Metal Ions

By far the most likely candidates for the other components with which H_2O_2 reacts are transition metal ions. Traces of metal ions as low as 10^{-9}M have been shown to affect peroxide stability (59). Such reactions generally produce hydroxyl and/or perhydroxyl radicals (39), which are known to react with MBG (52). The addition of such metal ions to the oxygen-alkali-MBG system brought about faster initiation of the degradation. When a small amount of ferric ion was added to an alkaline electrolyte containing H_2O_2, catalysis was so prevalent that the polarogram consisted only of a catalytic wave; the normal H_2O_2 reduction wave was eliminated entirely. The catalyzed hydrogen peroxide decomposition is probably an important source of free radicals under oxygen bleaching conditions, even when steps are taken to minimize metal ion contamination.

The only hypothesis which is consistent with all the data involves deactivation of catalytic metal ions by magnesium, to inhibit H_2O_2 decomposition. The ability of magnesium to remove metal ions from solution is well-documented (1,52, 60). Polarography demonstrated that magnesium tends to eliminate the catalyzed reduction wave of alkaline hydrogen peroxide, the wave which was enhanced by

ferric ion. Magnesium also eliminated small waves attributable to transition metals. The theory of a magnesium-metal ion interaction was also supported by the work of Gilbert, et al. (58).

As mentioned previously, it is conceivable that metal ions could produce initiating radicals by reacting with species other than H_2O_2. Such reactions could include

$$RO^- + M^{+n+1} \longrightarrow RO\cdot + M^{+n}, \qquad (1)$$

and

$$O_2 + M^{+n} \longrightarrow O_2^{\cdot -} + M^{+n+1}, \qquad (2)$$

where RO^- is an ionized carbohydrate hydroxyl group, and M is a transition metal. Neither of these reactions would be expected to be as important as H_2O_2 decomposition, however.

In this connection, Minor and Sanyer (49) noted that D-glucitol degradation did not occur when either oxygen or alkali was absent from the system. When oxygen was added to the system of sugar + alkali, an induction period of the usual length preceded the start of degradation. If a reaction such as (1) were important, it would produce initiating radicals prior to the introduction of oxygen. One would, therefore, not expect to observe the usual induction period after oxygen addition. Moreover, when alkali was added to the system of sugar + oxygen, the induction period again preceded the start of degradation. By similar reasoning, this provides evidence against Reaction (2). Furthermore, if the primary function of magnesium were to prevent metal ions from reacting as in (2), its excellent stabilizing ability with MBG in alkaline hydrogen peroxide (52) would be unexplained.

PROPOSED MECHANISM FOR OXYGEN-ALKALI DEGRADATION OF METHYL β-GLUCOSIDE

Production of a Keto Derivative

Most workers agree that the cellulose chain scission reaction in oxygen-alkali involves the oxidation of a C2 or C3 hydroxyl group, to produce a ketone (11,13-16, 20,25,27,29,61). This carbonyl derivative then undergoes rapid reaction with alkali, to yield new acidic end groups (14,15,20,21). As pointed out by Brooks (61), the rate of production of keto groups undoubtedly determines the rate of chain cleavage. The mechanism of this oxidation of secondary hydroxyl groups has remained a mystery, however.

The information gained in the present research, in conjunction with results of other workers, has allowed the postulation of a series of reactions which may be important during MBG degradation:

$$G + OH^- \underset{}{\overset{K_{12}}{\rightleftharpoons}} G^- + H_2O \qquad (12)$$

$$G^- + O_2 \xrightarrow{k_{13}} Z + OOH^- \qquad (13)$$

$$OOH^- + M^{+n} + H_2O \underset{k_{-14}}{\overset{k_{14}}{\rightleftharpoons}} \cdot OH + M^{+n+1} \ (+ 2OH^-) \qquad (14)$$

$$OOH^- + M^{+n+1} \underset{k_{-15}}{\overset{k_{15}}{\rightleftharpoons}} O_2^{\cdot-} + M^{+n} \ (+ H^+) \qquad (15)$$

$$\cdot OH + G \xrightarrow{k_{16}} R\cdot + H_2O \qquad (16)$$

$$R\cdot + O_2 \xrightarrow{k_{17}} Z + O_2^{\cdot-} \ (+ H^+) \qquad (17)$$

$$O_2^{\cdot-} + H_2O \xrightarrow{k_{18}} \cdot OH + OOH^- \qquad (18)$$

$$O_2^{\cdot-} + M^{+n+1} \xrightarrow{k_{19}} O_2 + M^{+n} \qquad (19)$$

$$\cdot OH + M^{+n} \xrightarrow{k_{20}} OH^- + M^{+n+1} \qquad (20)$$

$$2 \cdot OH \xrightarrow{k_{21}} OOH^- \ (+ H^+) \qquad (21)$$

$\underline{G}$ = a reactive carbinol group $H-\overset{|}{\underset{|}{C}}-OH$ of the MBG molecule

$\underline{G}^- = H-\overset{|}{\underset{|}{C}}-O^-$ $\quad \underline{Z} = \diagdown C{=}O \diagup$ $\quad \underline{R}\cdot = -\overset{|}{\underset{|}{C}}-OH$ $\quad \underline{M}$ = catalytic metal

If one makes the steady-state approximations that the concentrations of perhydroxyl ion, each species of metal ion, and all radicals do not change appreciably with time, the following expression for reaction rate is obtained:

$$d[Z]/dt = K_{12}k_{13}[G][OH^-][O_2] + \{[G]^2(b-1) - 2a[G] + [G]X^{\frac{1}{2}}\}/4c, \qquad (22)$$

where

$$\underline{X} = \{[\underline{G}](\underline{b}-1) - 2\underline{a}\}^2 + 8\underline{K}_{12}\underline{k}_{13}\underline{c}(\underline{b}+1)[\underline{G}][OH^-][O_2],$$

$$\underline{a} = \underline{k}_{20}[\underline{M}^{+n}]/\underline{k}_{16},$$

$$\underline{b} = \underline{k}_{18}/\underline{k}_{19}[\underline{M}^{+n+1}], \text{ and}$$

$$\underline{c} = \underline{k}_{21}/\underline{k}_{16}{}^2.$$

When experimental data are substituted into this equation, values for $(\underline{K}_{12}\underline{k}_{13})$, $\underline{a}$, $\underline{b}$, and $\underline{c}$ can be obtained. Thus, Equation (22) becomes, at $[OH^-] = 1.25\underline{N}$.

$$d[Z]/dt = 7.56 \times 10^{-5}P_{O_2}[G] + [G]\{(10.19[G] - 0.1870) + Y^{\frac{1}{2}}\}/16.8 \qquad (23)$$

where

$$\underline{Y} = (10.19[\underline{G}] - 0.1870)^2 + 3.084 \times 10^{-2}\underline{P}_{O_2}[\underline{G}],$$

and $\underline{P}_{O_2}$ is the partial pressure of oxygen on the system, in p.s.i. at 25°C. The derivation of Equations (22) and (23) is given in Appendix V, along with a discussion of the fit between Equation (23) and experimental kinetic data. The closeness of this fit lends credence to the postulated mechanism.

In the proposed scheme, Reactions (12) and (13) would be the only important ones during the induction period. As the peroxide concentration builds up from (13), the initiating Reactions (14) and (15) would produce more and more free radicals. The radical chain reaction of glucoside is propagated primarily by (16), (17), and (18). Reactions (19), (20), and (21), and the reverse of (14) and (15), bring about chain termination.

As seen in Fig. 3, the steady-state assumption for peroxide ($d[OOH^-]/dt = 0$) is a rather poor approximation during the first few hours of reaction. For a brief nonsteady-state time near the end of the induction period, the production of radicals by Reactions (14) and (15) would be more rapid than at equilibrium. One would, therefore, expect the reaction rate during this time to depend on concentrations of peroxide and catalytic ions. As equilibrium is achieved, however, this approximation and, hence, Equation (23) become more valid. At equilibrium, then, the reaction rate should become independent of peroxide and metal ion concentrations; experimentally, this was observed (Fig. 2, 3, 5, and 6).

The presence of magnesium would reduce the effective metal ion content, and so retard the production of initiating radicals by (14) and (15). But again, such a reduction in catalytic ion activity would not change the postinduction period reaction rate. This was experimentally found to be true in the MBG system (Fig. 2 and 3). The addition of excess iodide ion would allow Reactions (4) and (6) to compete effectively with Reactions (14) and (15). Thus, the only glucoside degradation which would occur in this case would be by Reaction (13) (cf. Fig. 5 and 6).

The only peroxide produced in the proposed reaction scheme is the hydrogen peroxide intermediate. Presumably "other peroxides" would be produced by side reactions, such as

$$2\ RO\cdot \longrightarrow ROOR \qquad (24)$$

or

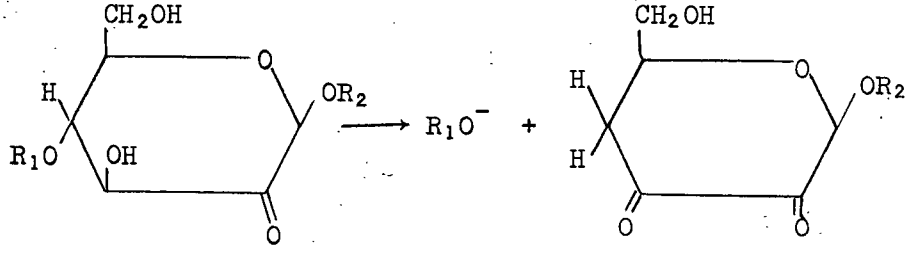
$$RO\cdot + \cdot OH \longrightarrow ROOH. \qquad (25)$$

Small amounts of RO· could be produced by Reaction (1), for example. Additional sources of "other peroxides" could include reactions of primary oxidation products (hydroxy acids) with either oxygen, perhydroxyl ions, or free radicals.

Subsequent Reactions of the Keto Derivative

In the case of MBG, the intermediate Z could be a 2-, 3-, or 4-keto derivative of the glucoside. As mentioned above, Z would be very unstable in hot alkali (14, 15,20,21), and may undergo some of the reactions depicted in Fig. 20. These proposed pathways seem to be in line with the still meager data available concerning reaction products. Undoubtedly, some of these reactions are much less important than others. The final products indicated can probably undergo subsequent reactions in the system; the only one isolated and identified in MBG reaction solutions is the methyl 2-carboxypentofuranoside (21).

Obviously, if the carbohydrate had a 4-O-substituent, it could be eliminated by the same mechanism proposed here for the methyl group of MBG. For the case of a glucose unit in the middle of a cellulose chain, the following could occur:



(26)

R_1O^- is a shortened cellulose chain with a new glucose end group. It would be rapidly oxidized, probably to produce hydrogen peroxide and an aldonic acid, as discussed in the following section.

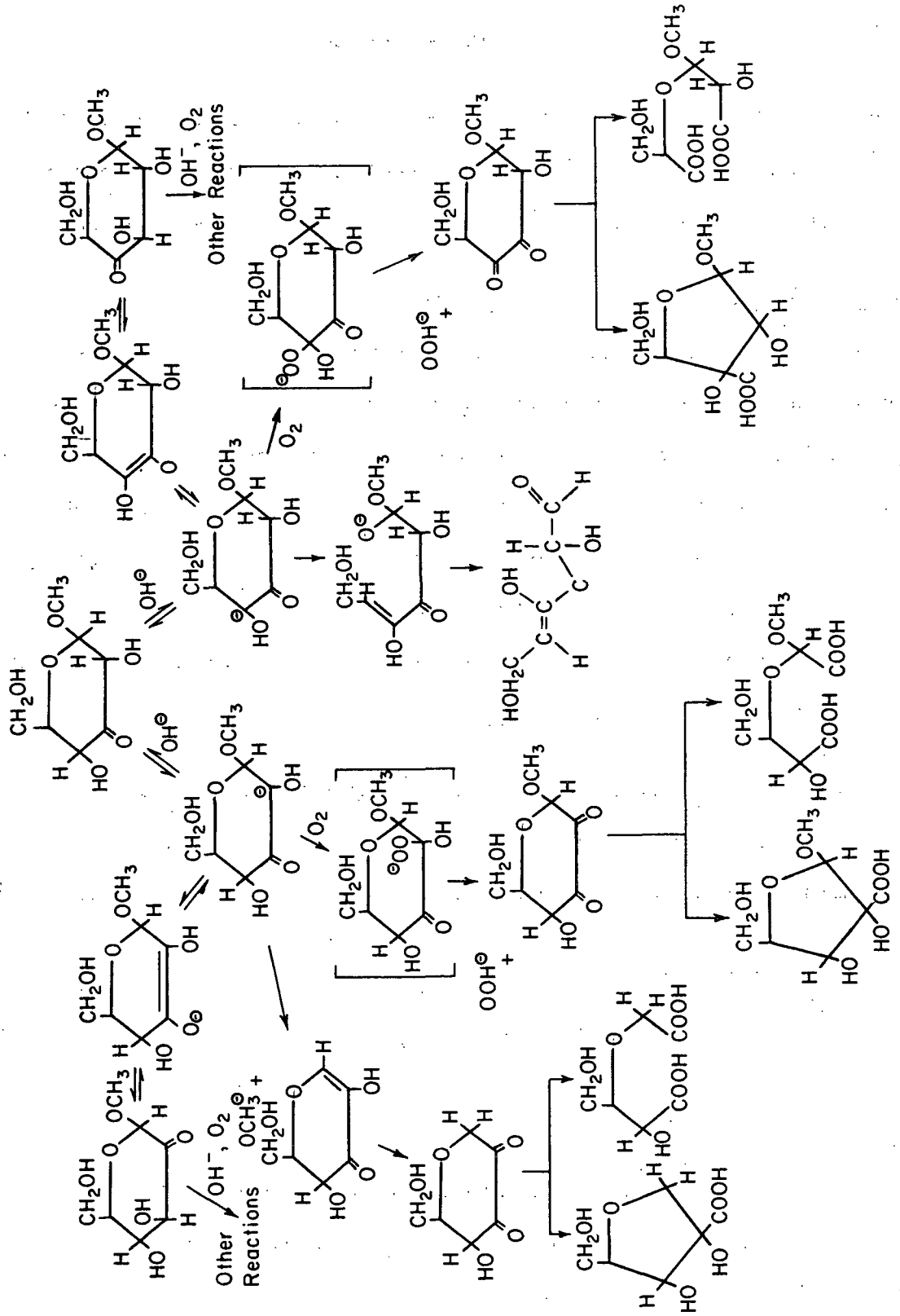


Figure 20. Possible Reactions of a Keto Derivative of MBG in Oxygen-Alkali

MECHANISM OF OXYGEN-ALKALI DEGRADATION OF D-GLUCOSE

The finding that H_2O_2 is the only stable peroxide produced from glucose is significant. It explains more precisely why a reducing sugar accelerates the degradation of a nonreducing sugar in oxygen-alkali. Thus, MBG reacts more rapidly in the presence of glucose (11), undoubtedly because of the rapid production of hydrogen peroxide from the reducing sugar.

Bamford and Collins (36) and DeWilt and Kuster (37) proposed that relatively stable organic peroxides were produced as intermediates in the glucose reaction. The fact that H_2O_2 is the only detectible peroxide produced serves to clarify the ionic degradation mechanism. On the basis of this information and that reported by other authors (33-37,40,62), the reactions in Fig. 21 are postulated to occur in excess oxygen and alkali at room temperature. Under other conditions, competing reactions (formation of other aldonic and saccharinic acids) may become important (33,36). At higher temperatures, free radical reactions may complicate the picture (35).

The pathway at the top of Fig. 21 yields C1-C2 cleavage products without producing a stable peroxide. This pathway, proposed by several authors (36,37, 40,62), is consistent with the experimental observation that the rate of H_2O_2 production is less than the pseudo first-order rate of glucose degradation.

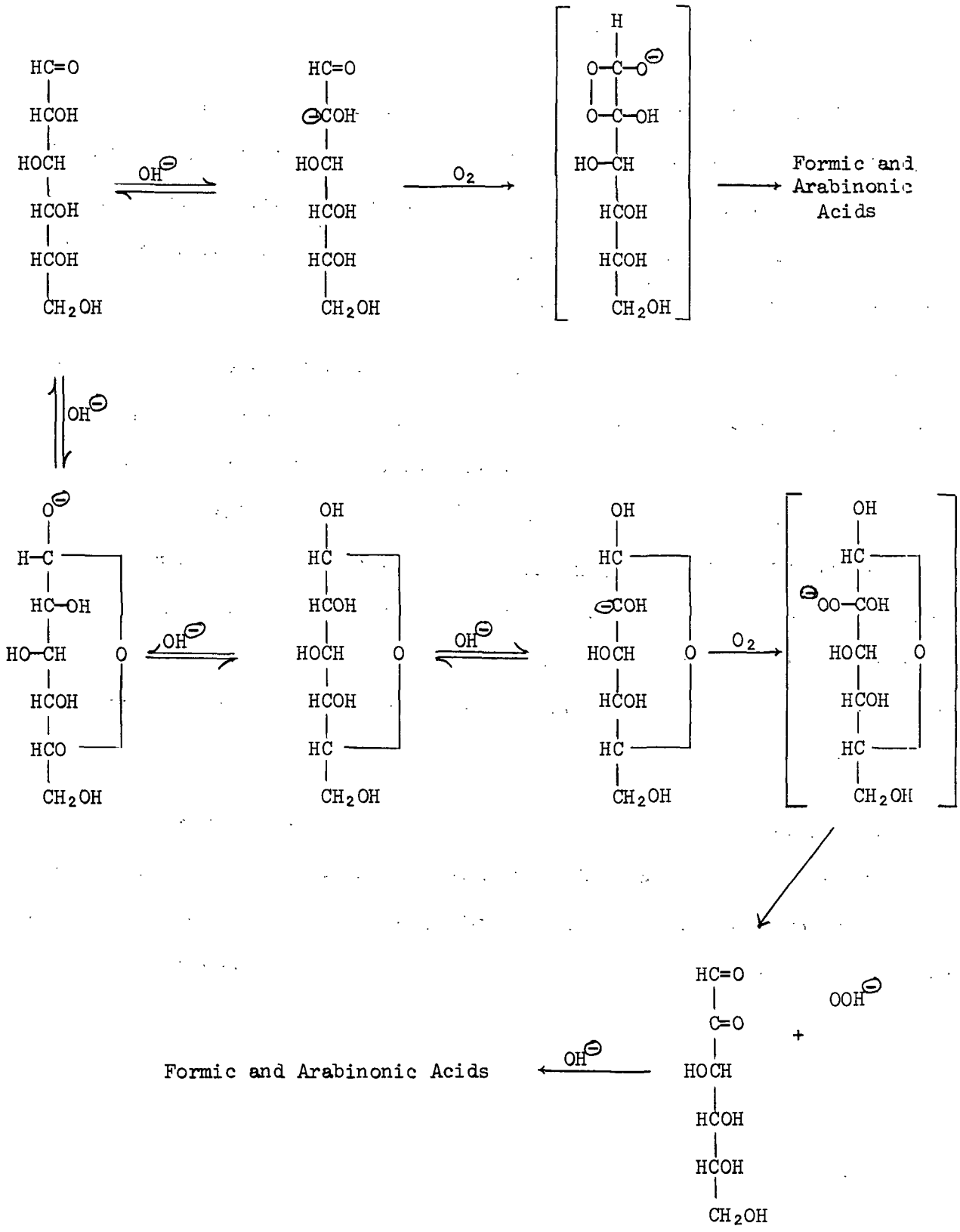


Figure 21. Proposed Pathways in the Room Temperature Reaction of D-Glucose in Strong Alkali and Excess Oxygen

CONCLUSIONS

When MBG is allowed to react in oxygen-alkali at 120°C., hydrogen peroxide is produced as an intermediate. Other more stable peroxides are produced as by-products.

Based on the observed effects of ferric, magnesium, and iodide ions on the degradation of MBG and on the stability of the H_2O_2 intermediate, it is concluded that the trace metal ion-catalyzed decomposition of hydrogen peroxide produces free radicals which initiate carbohydrate degradation. Results of MBG reactions, and polarographic work, led to the further conclusion that magnesium stabilizes the H_2O_2 intermediate by deactivating catalytic ions. This peroxide stabilization effects the lengthening of an otherwise very brief induction period, or the postponement of the start of a period of more rapid glucoside degradation. Iodide ion also lengthens the induction period, presumably because of its ability to reduce H_2O_2 by an ionic mechanism. This ionic reaction effectively competes with the radical-producing peroxide decomposition which is caused by trace catalytic ions.

A tentative mechanism for the degradation of MBG is proposed. Hydrogen peroxide, which builds up during the induction period, is postulated to react with trace catalysts to produce the radicals $\cdot OH$ and $O_2^{\cdot-}$. These radicals are then involved in the propagation of a chain reaction, whereby a secondary hydroxyl group of MBG is oxidized to a carbonyl. This keto derivative rapidly reacts with alkali, to produce a mixture of acidic degradation products. Termination of the chain reaction may occur by recombination of radicals, or by reaction of a radical with a multivalent metal ion. Other peroxides could be produced by side reactions, such as radical recombinations, or further reactions of primary oxidation products. A rate equation derived from this proposed reaction scheme fits the experimental data very well.

The ability of glucose to temporarily accelerate the degradation of a non-reducing sugar is attributed to a rapid production of hydrogen peroxide, the only stable peroxide produced from this reducing sugar.

SUGGESTIONS FOR FUTURE RESEARCH

This research has provided much insight into the mechanism of stabilization of a model carbohydrate in oxygen-alkali. The work now needs to be extended to more complex systems. For instance, the effects of a reducing sugar on the chain scission reaction have already been noted. It is likely that other components present in pulp would also have profound effects on carbohydrate degradation. Lignin, for example, would be a potential source of peroxides and free radicals. The effectiveness of magnesium in the presence of such compounds should be studied further.

It would also be useful to determine which species of metal ions are most destructive, and in what form they exist as catalysts (complexed, in colloidal dispersion, or precipitated as hydroxides). Another topic of practical importance involves the rate of transfer of oxygen to the reactive sites in a pulp slurry.

Attempts should be made to prove or disprove the proposed mechanism of glucoside degradation. Such studies might involve more extensive and precise kinetic work, product analysis, or investigation of other model compounds. A significant step would be made if one could conclusively demonstrate the presence of free radicals in the system. The author knows of no simple way to do this, however, especially at elevated temperatures in the presence of molecular oxygen.

In practical oxygen bleaching, the reaction may be stopped long before the system reaches equilibrium. That is, the bleach may be completed before the steady-state assumption for peroxide ever becomes valid. From this standpoint, then, it would be more useful to study the factors which influence degradation during this initial nonequilibrium period. As noted previously, it is only during

this initial period that metal ions and magnesium show any significant influence on MBG degradation.

In addition, a host of other questions must be answered before an adequate understanding of the oxygen-alkali system can be gained. A greater insight into the relevant chemical and physical phenomena would permit better control of oxygen and peroxide bleaching and alkali cellulose aging. It would also allow more intelligent evaluations of such related processes as oxygen and peroxide pulping.

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Finally, a special acknowledgment must be made to the One Who controls oxygen-alkali reactions (Colossians 1:16,17), and Whose guidance, when heeded, invariably proved to be perfect (Proverbs 3:5,6).

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APPENDIX I

CONSTRUCTION AND USE OF APPARATUS

OXYGEN REACTION APPARATUS

CONSTRUCTION OF REACTOR AND ACCOMPANYING EQUIPMENT

The basic reaction system was patterned after a design conceived by McCloskey (11). Details of reactor construction are illustrated in Fig. 22.

The stainless steel container was a one-liter Parr bomb 236HC10 cylinder.[3] The insert was machined from a solid teflon cylinder. The brass cover, bolted to the pot by means of flange fittings, was lined with teflon underneath. At first, this lining consisted of a cast film applied by commercial teflon applicators.[4] After several heating cycles, however, this coating started to develop checks and cracks; so to avoid a potential source of metal contamination from the cover, a 1/16-inch teflon disk was attached as shown in Fig. 22. The contact between the teflon cover lining and the insert provided an effective seal.

Figure 23 illustrates the design of the two ports in the reactor cover. The stainless steel connectors were Gyrolock 2COS-316 fittings[5]. The design of these fittings, and the snugness of the fit between the teflon tubing and the cover plate, effectively prevented gas leaks around the tubing. In the case of the port leading to the cooling coil and valve system (Fig. 23), the teflon tubing was one continuous piece, about 7-ft. long, extending from the bottom of the reactor through the cooling coil.

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[3] Parr Instrument Co., 211 Fifty-third St., Moline, Illinois.

[4] Ornamental Plastics, Sheboygan, Wisconsin.

[5] Hoke Mfg. Co., Box 501, Tenafly, New Jersey.

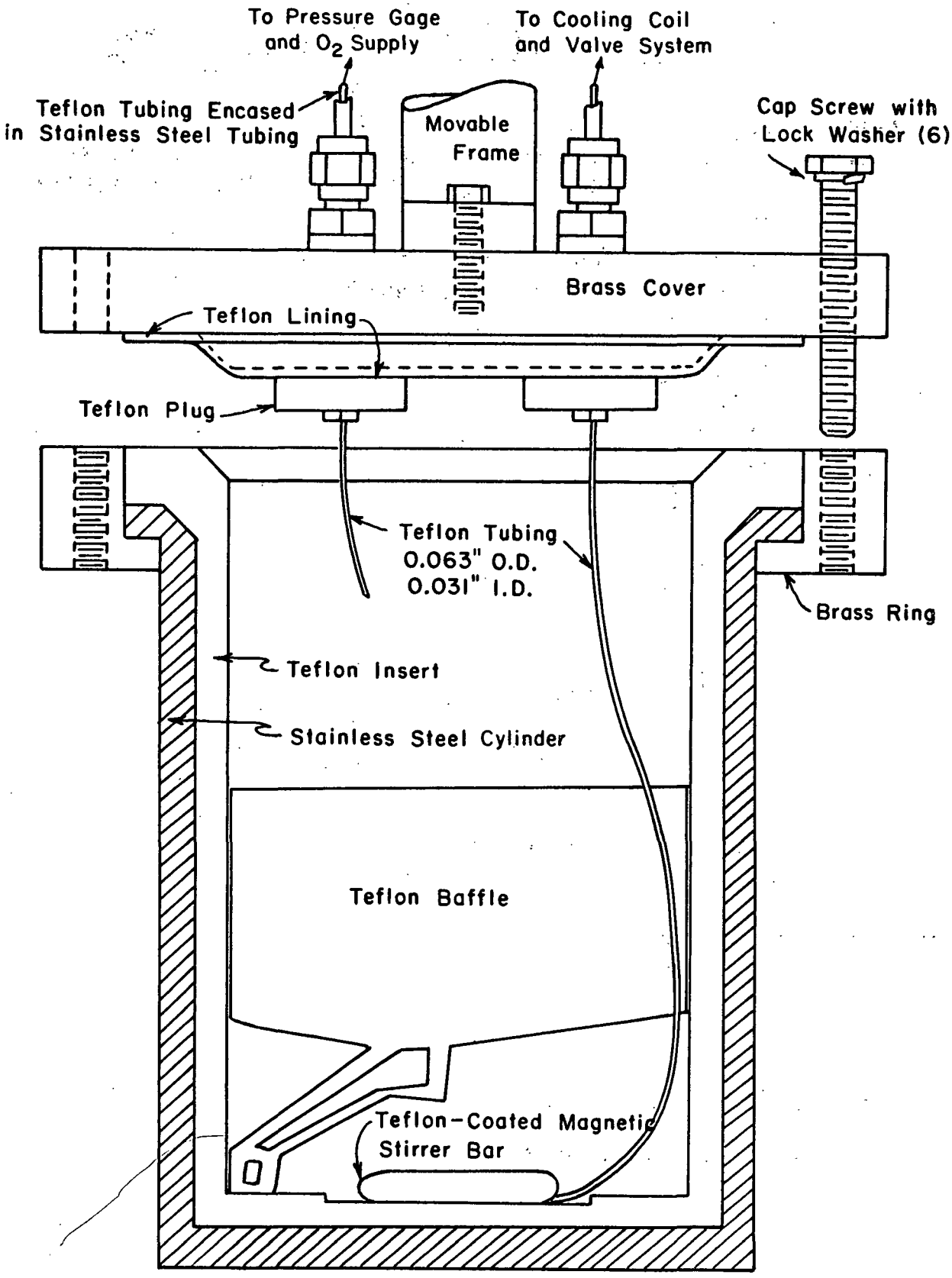

Figure 22. Reactor and Cover

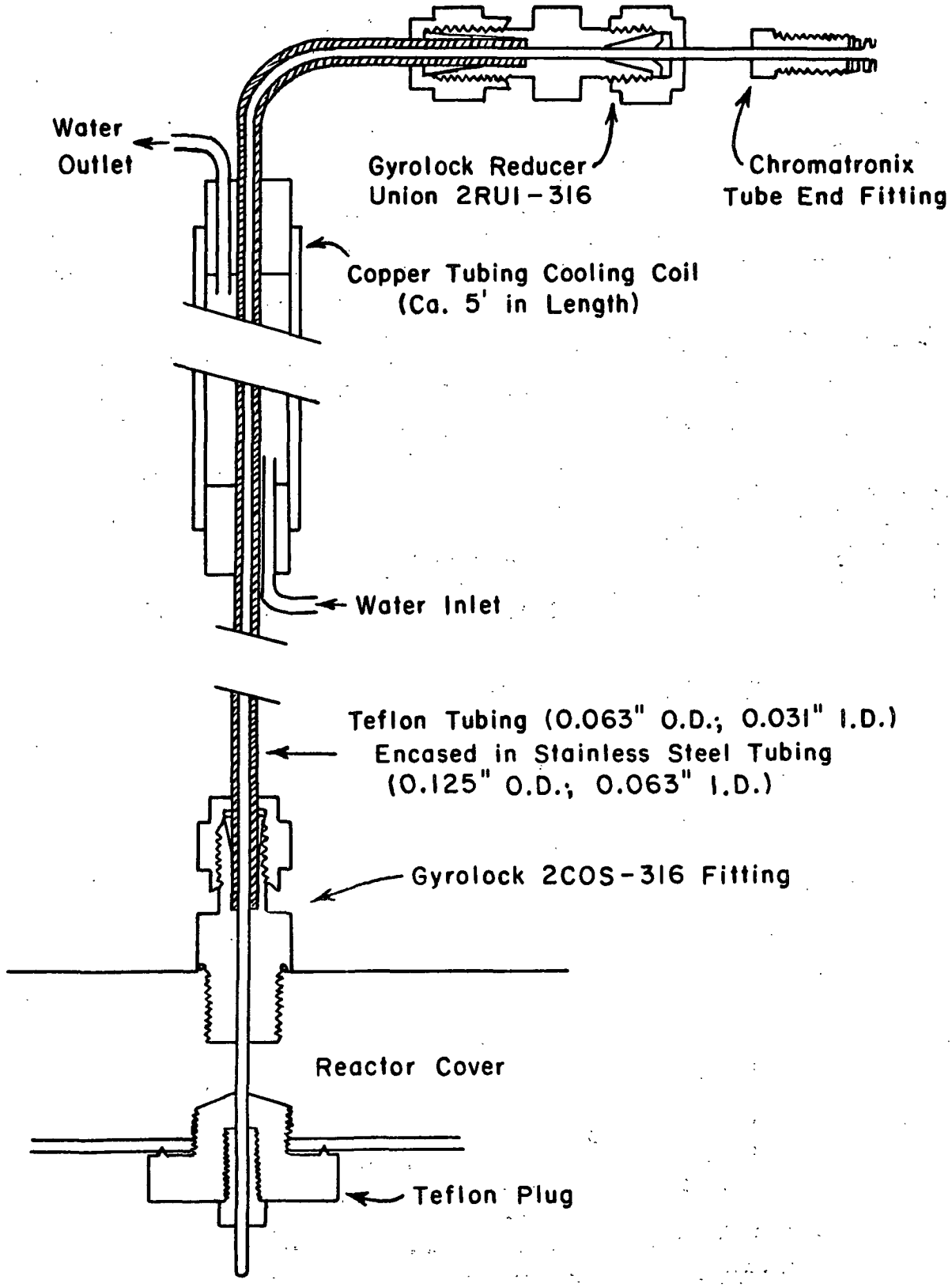

Figure 23. Sampling Tube Port and Connections

Teflon tubing could not retain its shape under the heat and pressure outside the reactor unless it were reinforced. Thus, it was necessary to back it with a snugly-fitting sheath of stainless steel tubing, from the cover through the cooling coil. When cooled to room temperature, the teflon tubing can withstand pressures far in excess of those encountered here, without reinforcement. On the cool end of the coil, the Gyrolock reducer union provided a final seal against any gas which may have escaped between the stainless steel and teflon tubings.

Figure 24 illustrates the construction of the apparatus accompanying the reactor. The cover was bolted to a T-frame which could be hung on brackets on the main frame when not in use. Also attached to the T-frame were the cooling coil, and the pressure gage-rupture disk-relief valve assembly. The reactor itself rested on the magnetic stirrer, which in turn was supported by another frame capable of being moved up and down with the counterweight. When the reactor was moved up and bolted to the cover, and the T-frame was removed from its brackets on the main frame, the entire assembly could be lowered so that the reactor was immersed in the oil bath.

The bursting pressure of the rupture disk was rated at 176 p.s.i.g. No rupture disk failures were encountered in the work.

The magnetic stirrer was an air-driven "Mag-Jet"[6]. It was modified by replacing its two small magnets with a larger 1½-inch cylindrical magnet. This modification resulted in much better tracking of the stirrer bar inside the reactor. This, combined with the use of the baffle shown in Fig. 22, provided for good dispersion of the vapor and liquid phases in the reactor.

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[6]Matheson Scientific, Inc., 1850 Greenleaf Ave., Elk Grove Village, Illinois.

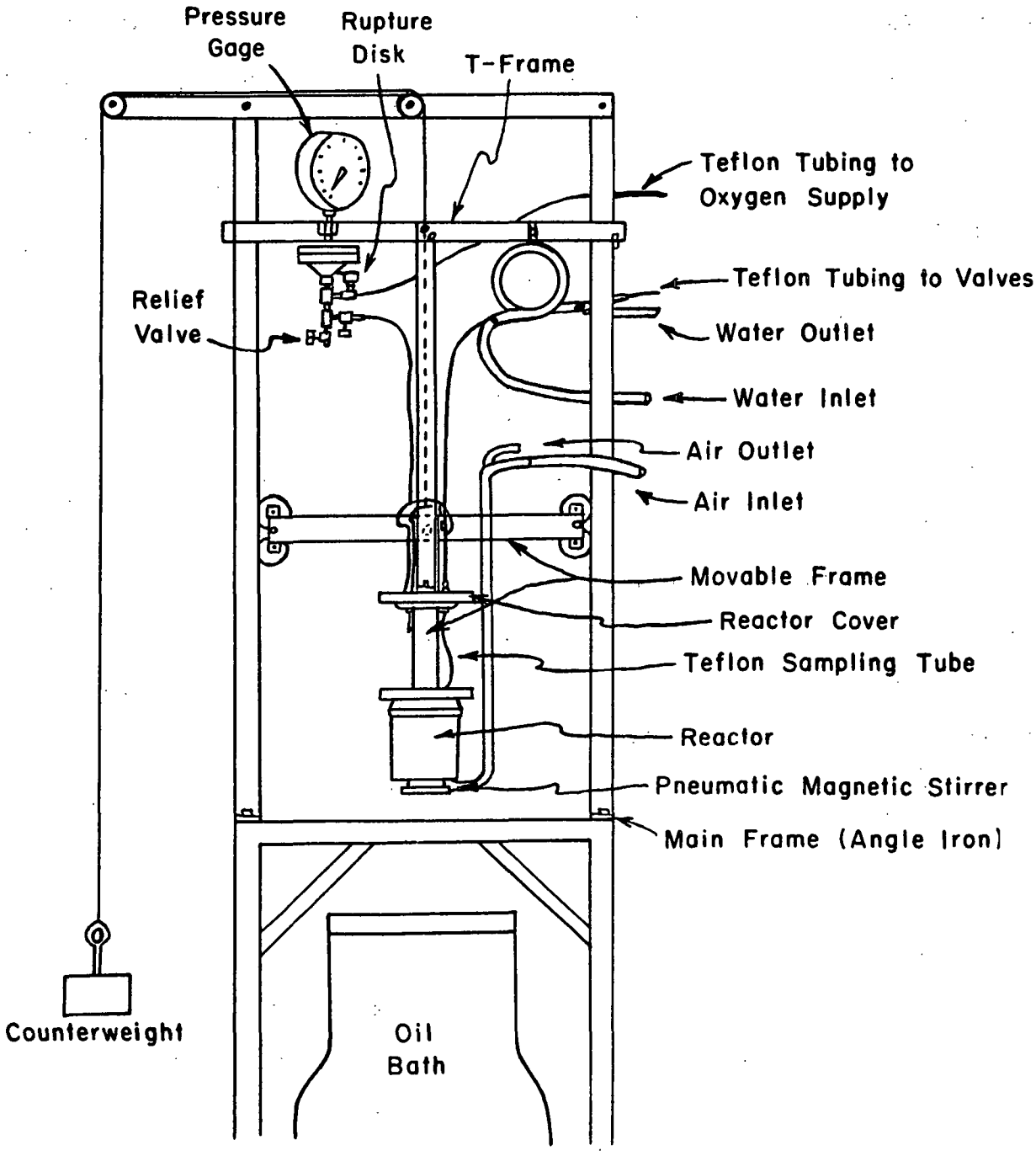

Figure 24. Apparatus for Oxygen-Alkali Reactions

The oil bath, two feet high and one foot in diameter, was constructed several years ago by E. F. Elton (64). It was equipped with twelve 350-watt strip heaters, and an immersion thermostat. This 220-volt bath heating circuit was used only to bring the bath fluid up to temperature initially. Finer temperature control (±0.3°C.) was achieved with an auxiliary 125-watt knife heater, and a control 250-watt knife heater. Both knife heaters operated through a CRC relay activated by a CRC thermoregulator[7]. High temperature polyalkylene glycol bath fluid[8] was used as the heating medium. The fluid was agitated with a 1/30 hp. Lightnin' Mixer driving a 2-foot shaft with two 3-inch propellers.

Figure 25 is a schematic depiction of the valve system used in connection with the reactor. All tubing connections, and Valves V1 through V4, were obtained from Chromatronix, Inc.[9] Valve V5 was a Hamilton on-off valve[10]. Valves V6 and V7 were ¼-inch stainless steel needle valves.

The line proceeding from the cooling coil was attached to a 2-foot length of small I.D. (0.016 inch) tubing (L1), to slow down the flow rate during sampling, and allow sufficient time for cooling the hot sample. The line then proceeded through on-off Valve V1, to Port 1 of the two-position valve V2. In one position, Ports 1 and 4 were connected; in the other position, Port 1 was connected to 2, and 3 was connected to 4. Coil L2 consisted of larger I.D. tubing, with a total volume of ca. 33 ml. The top end of this coil was connected to the Central Port C of Rotary Valve V3. This valve allowed the central port to be connected to any one of the Peripheral Ports 1-6. Valve V4 allowed either oxygen or nitrogen to be

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[7]Chemical Rubber Co., 2310 Superior Ave., Cleveland, Ohio.

[8]Matheson Scientific, Inc., 1850 Greenleaf Ave., Elk Grove Village, Illinois.

[9]Chromatronix, Inc., 2743 Ninth St., Berkeley, California.

[10]The Hamilton Co., Whittier, California.

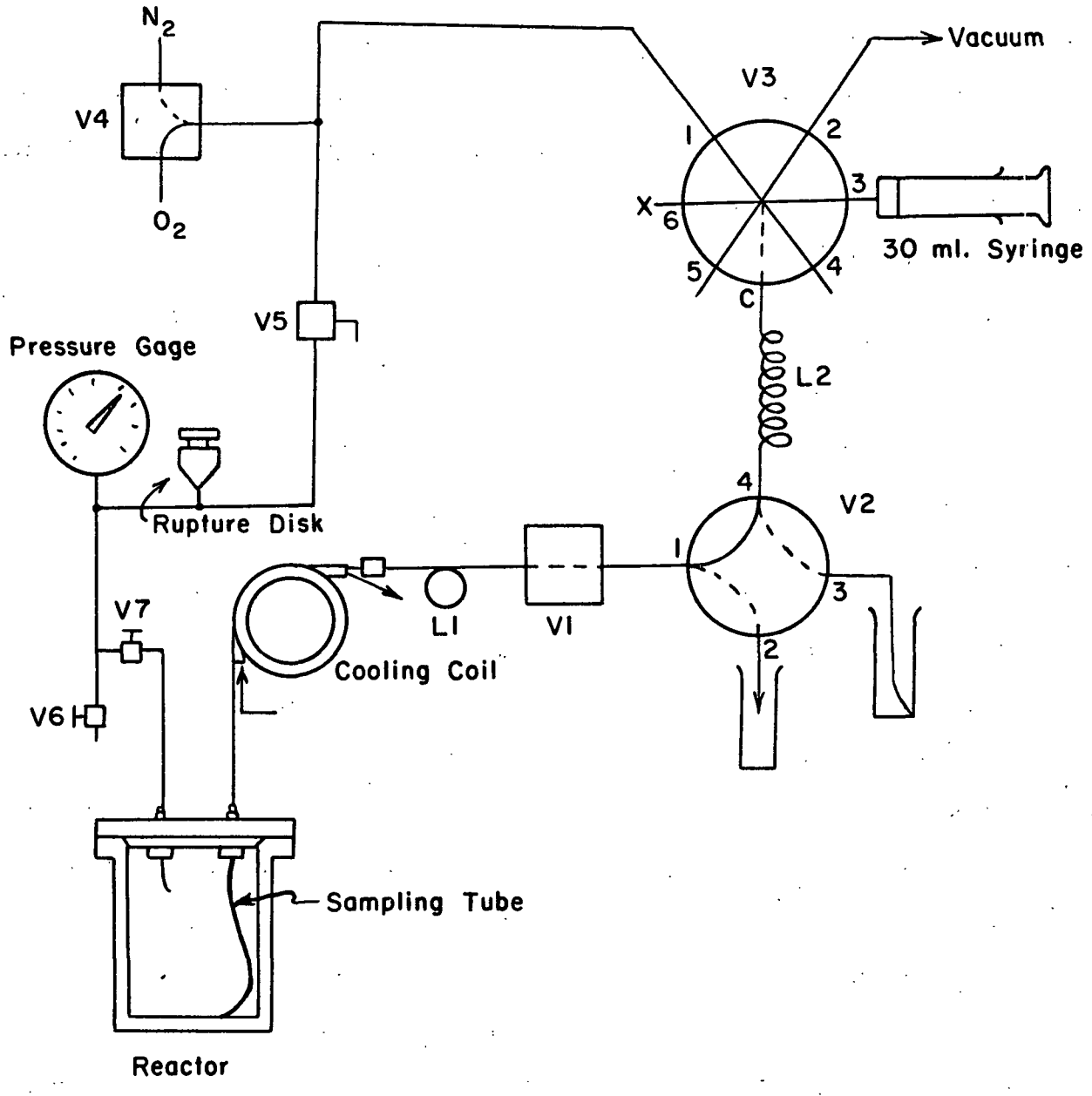

Figure 25. Valve System

forced through the valve system, or through the pressure gage line into the reactor. The operation of the valve system is discussed in the next section.

OPERATION OF APPARATUS

Prior to use, the reactor, stirrer bar, and baffle were washed according to the same procedure described later for glassware. The valve system and lines were washed twice with 5% hydrochloric acid, then several times with triply-distilled water. These liquids were moved through the lines by using the syringe and nitrogen pressure. The reactor was allowed to air-dry, and the lines were dried by application of the vacuum for several minutes.

As soon as the solution was added to the reactor, the magnetic stirrer was turned on, and nitrogen was passed through the sampling tube, via V3, V2, and V1. After the cover was bolted on, nitrogen purging was continued for several minutes more, with the gas escaping from Relief Valve V6. Then the nitrogen was shut off, and Valves V1, V6, and V7 were closed.

The oil bath temperature was allowed to overshoot to 130°C. before the reactor was immersed, to minimize heat-up time. At 30-45 minutes after immersion, the bath started controlling at 120°C. In preliminary experiments with the pressure gage open to the reactor during heat-up, the pressure leveled off at ca. 32 p.s.i.g. after about one hour. Thus, at least 90 minutes were allowed to attain temperature equilibrium in the reactor.

The reaction was started by opening the oxygen valve, then V5 and V7, to pressurize to the desired level. The desired gage pressure was calculated by adding the needed oxygen pressure, corrected to 120°C., to the 32 p.s.i.g. reading before pressurization. If it was desired to add a reagent, such as glucose or hydrogen peroxide, at the start of reaction, a different procedure was followed.

The reagent in solution was drawn from Port 3 of V2 into L2, using the syringe. Then V2 was switched to the 1-4 position, and the solution was forced into the reactor with oxygen, through V3, V2, and V1. Valve V1 was then closed, and oxygen was added to the desired level through V5 and V7.

During reaction, Valves V5 and V7 were open, and Valves V1 and V6 were closed. Valve V3 was set at a position between ports, and V2 was in the 1-2,3-4 position. When a sample was desired, Valve V1 was opened. At a gage pressure of ca. 130 p.s.i., the sample could be collected at Port 2 of V2 at a rate of about 10 ml./min. Peroxide analyses were immediately run on the samples, and aliquots were saved in polyethylene bottles for later glucoside analyses.

POLAROGRAPH AND ACCOMPANYING EQUIPMENT

DESCRIPTION OF EQUIPMENT

A Sargent Model XV Recording Polarograph and a Sargent D.M.E. assembly were obtained from Dr. D. G. Williams. The D.M.E. assembly provided a support for the mercury reservoir, the D.M.E., and the 40-ml. capacity cell. A schematic of the cell and the reference saturated calomel electrode (S.C.E.) is shown in Fig. 26. The D.M.E. was connected to the mercury reservoir by means of rubber tubing, with a pinch clamp to turn flow on and off. This cell and electrode assembly is similar to that used by O'Connor in his thesis (65).

The cell was fitted with a three-way stopcock, providing two inlets for water-saturated nitrogen. One inlet was for prepurging via a gas dispersion tube; the other was used to blanket the solution with nitrogen during recording.

Electrical connection between the S.C.E. and the electrolyte solution was made via a potassium chloride-agar salt bridge in a ¼-inch I.D. bent glass tube.

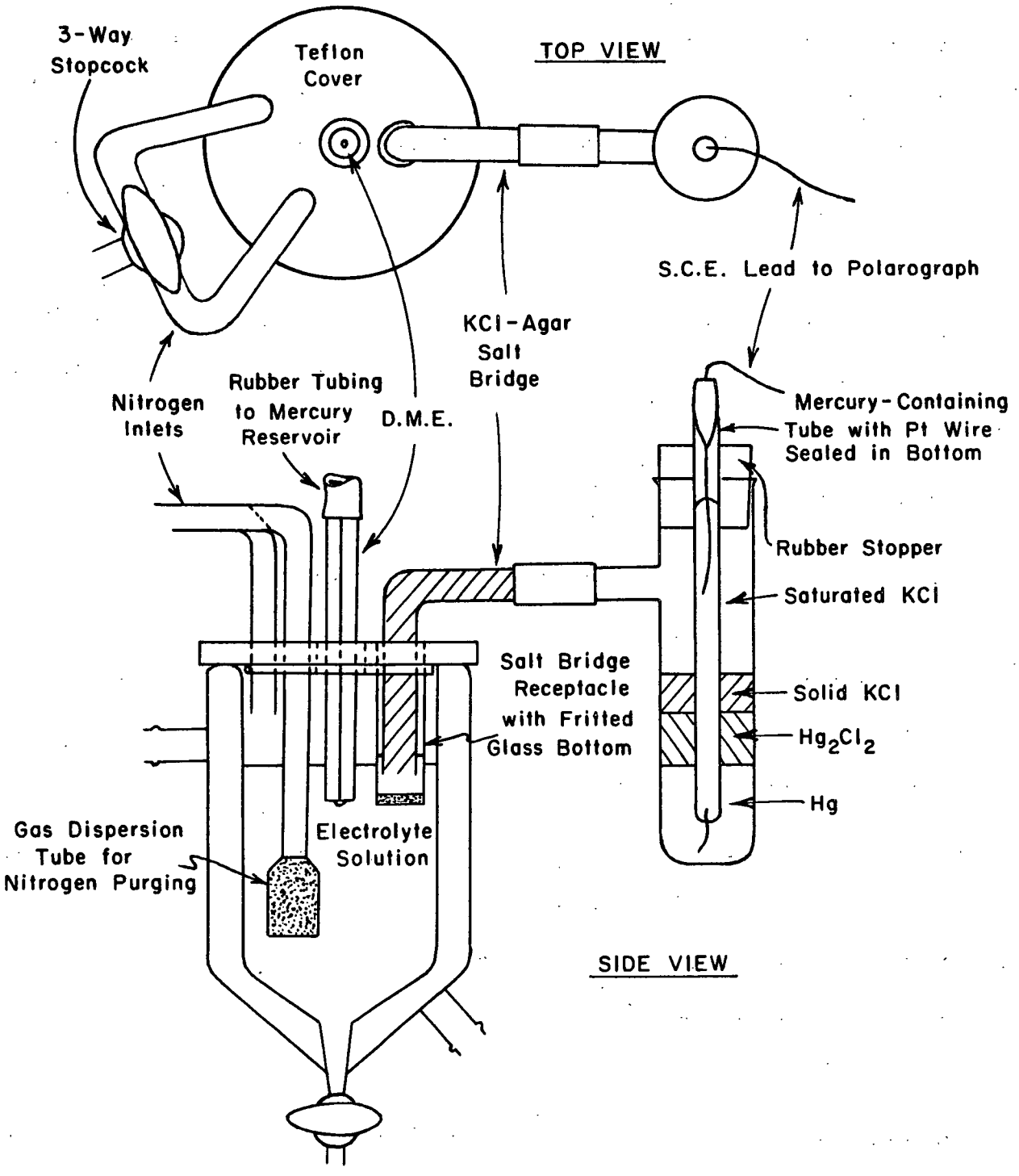

Figure 26. Polarographic Cell and Reference Electrode

The agar gel was prepared by heating a flask containing granular agar (4.0 g.) in water (90 ml.), in a boiling water bath. When solution was complete, potassium chloride (30.0 g.) was added. After this dissolved, the glass tube was filled with the solution, and it was allowed to cool and gel. The filled tube was then attached to the S.C.E. vessel. To prevent the gel from drying out, the exposed end of the salt bridge was kept immersed in saturated potassium chloride solution when not in use in the cell.

The S.C.E. was prepared as indicated in Fig. 26, in a 3-inch test tube with a side arm. To prevent the creeping of potassium chloride, the rubber stopper was coated liberally with silicone grease.

POLAROGRAPHIC PROCEDURE

The cell was washed with the electrolyte solution prior to filling. Unless it was desired to obtain a polarogram of dissolved oxygen, the solution was pre-purged with water-saturated nitrogen for 1-3 minutes, via the gas dispersion tube. The three-way stopcock was then switched to allow nitrogen to flow over the solution, the D.M.E. was immersed, and the polarogram was recorded.

After obtaining the polarogram, the D.M.E. was raised out of the cell, and rinsed with distilled water. The mercury which had dropped from the D.M.E. was removed from the bottom of the cell through the stopcock. After the cell was emptied, the gas dispersion tube, the salt bridge receptacle, and the cell were thoroughly rinsed with distilled water.

In order to prevent clogging of the D.M.E. capillary, it was never allowed to stand in a solution when the mercury was not flowing. After completing a series of polarograms, the mercury flow was kept on while the D.M.E. was dipped in 1:1 nitric acid, then rinsed thoroughly in a stream of distilled water from

a wash bottle. When the capillary tip had air dried, it was moved up just below the mercury reservoir, and the flow was stopped by clamping the rubber tubing.

The cell resistance was determined by recording polarograms of three widely different concentrations of cadmium sulfate in 1.25N potassium chloride electrolyte. The slope of a plot of half-wave potentials vs. diffusion currents provided the cell resistance (57).

In studying hydrogen peroxide, the residual curve could be obtained by recording the polarogram of the electrolyte with no peroxide. A solution of H_2O_2 (less than 1 ml.) was then added, and the electrolyte solution was stirred by forcing nitrogen through the gas dispersion tube. After recording this polarogram, a 10-ml. aliquot was pipetted out for colorimetric peroxide analysis. Additional peroxide could then be stirred in, the polarogram again recorded, and another peroxide analysis done. Thus, one sample of a given electrolyte could provide polarograms at two different peroxide concentrations. The dilution of the electrolyte caused by adding the peroxide was of little consequence.

Ten minutes were required to obtain a polarogram from 0 to -2 volts. During this time, peroxide decomposition was generally small.

APPENDIX II

MATERIALS AND REAGENTS

METHYL β-D-GLUCOPYRANOSIDE

MBG was obtained from J. T. McCloskey. It was repurified by refluxing in 5% sodium hydroxide, followed by deionization and recrystallization from ethanol (3X). The melting point range, determined in a Thomas-Hoover capillary apparatus, was 110-111°C. The optical rotation, obtained using a Zeiss-Winkel polarimeter, was $[\alpha]_D^{21} = -33°$.

WATER

All water used for preparing solutions and for cleaning apparatus was triply-distilled. Water from a distilled water tap was dripped into a one-liter pot containing boiling alkaline permanganate (0.2 g. $KMnO_4$/liter; 0.5 g. NaOH/liter). The condensate from this pot dripped into a second flask containing boiling sulfuric acid solution (50 ml. concd. sulfuric acid/liter). The final condensate was collected and stored in a pyrex bottle.

SODIUM HYDROXIDE

Stock solutions of sodium hydroxide were prepared and purified to minimize trace contamination. Reagent-grade pellets (ca. 450 g.) were dissolved in freshly distilled water (500 ml.), and allowed to cool. A solution (50 ml.) of equal volumes of ethanol and water, containing phenyl 2-pyridyl ketoxime (100 mg.), was added to the alkali solution with stirring. This reagent forms alkali-soluble colored complexes with a variety of metal ions (63). The complexes were removed by extracting with isoamyl alcohol (100 ml.) and chloroform (2 × 100 ml.). Residual chloroform was removed by heating at 70-80°C. As an added precaution

against organic matter, 30% hydrogen peroxide (2-3 ml.) was stirred into the solution. After standing for at least one day, the solution was heated to boiling to destroy the peroxide, and filtered through a medium-porosity glass filter to remove any carbonate precipitate. The stock solution was stored in a pyrex bottle under a nitrogen atmosphere. Each time a sample was pipetted from this bottle, the solution was again purged with nitrogen before sealing. Using this procedure, no carbonate precipitate was observed in the solution even after several months.

The stock sodium hydroxide solution was standardized by titrating it against known amounts of primary standard potassium acid phthalate, to a bromcresol purple end point.

SODIUM THIOSULFATE

To the fresh triply-distilled water (3 liters) were added sodium thiosulfate pentahydrate (7.50 g.) and sodium carbonate (0.3 g.). The solution was shaken and allowed to stand at least one day. It was then standardized by titrating against a standard solution of potassium iodate (0.0712 g. KIO_3/liter), to starch end point.

OTHER REAGENTS

Matheson Extra Dry Grade[11] oxygen was used. It is supposedly at least 99.6% pure, with a dew point below -70°F. Nitrogen was Matheson prepurified[11].

The phenyl 2-pyridyl ketoxime was obtained from Eastern Chemical Corp.[12] The titanium sulfate was a purified grade of $TiOSO_4 \cdot H_2SO_4 \cdot 8H_2O$ from Fisher[13]. All other chemicals used were reagent grade.

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[11] The Matheson Company, P.O. Box 960, Joliet, Illinois.

[12] Eastern Chemical Corporation, P.O. Box H, Pequannock, New Jersey.

[13] Fisher Scientific Co., 1458 N. Lamon Ave., Chicago, Illinois.

GLASSWARE

Precautions were taken to minimize impurities introduced into the solutions from the walls of glassware. All glassware used in peroxide stability studies and in preparation of reaction solutions was first washed successively with 5% sodium hydroxide, water, 5% hydrochloric acid at ca. 60°C., 5% nitric acid at ca. 60°C., and finally several times with triply-distilled water.

APPENDIX III

ANALYTICAL PROCEDURES

ANALYSIS OF PEROXIDES

Peroxide determinations were carried out using a modified colorimetric method which employs acidic titanium sulfate (41-45). A straight-line calibration curve relating peroxide concentration to absorbance (Fig. 27) was obtained by analyzing solutions of various hydrogen peroxide concentrations, both by iodide-thiosulfate titration (66), and by the titanium sulfate colorimetric method.

The thiosulfate titrations were carried out as follows: The sample (25 ml.) was pipetted into a 250-ml. Erlenmeyer flask containing 5N sulfuric acid (20 ml.). Sodium carbonate (ca. 0.2 g.) was added with swirling, to purge out dissolved oxygen. Ammonium molybdate (ca. 0.2 g.) and 1% potassium iodide solution (50 ml.) were then added, and the solution was titrated against standardized sodium thiosulfate solution (0.00929N). The peroxide concentration was calculated from Equation (27):

$$[\text{Peroxide}] = V_t C_t / 2V_s, \qquad (27)$$

where V_t and C_t are the volume and concentration of the thiosulfate solution, respectively, and V_s is the volume of original sample.

Colorimetric peroxide analyses were carried out by the following procedure: to a 50-ml. volumetric flask was added 5N sulfuric acid. The amount used (10-13 ml., depending on the pH of the sample) was sufficient to give a final acid concentration of ca. 1N. The sample (10 ml.) was then added to this, followed by 1 ml. of titanium sulfate solution (10 g. $TiOSO_4 \cdot H_2SO_4 \cdot 8H_2O$ and 10 ml. concd. H_2SO_4 diluted to 100 ml.). The test solution was diluted to the 50-ml. mark,

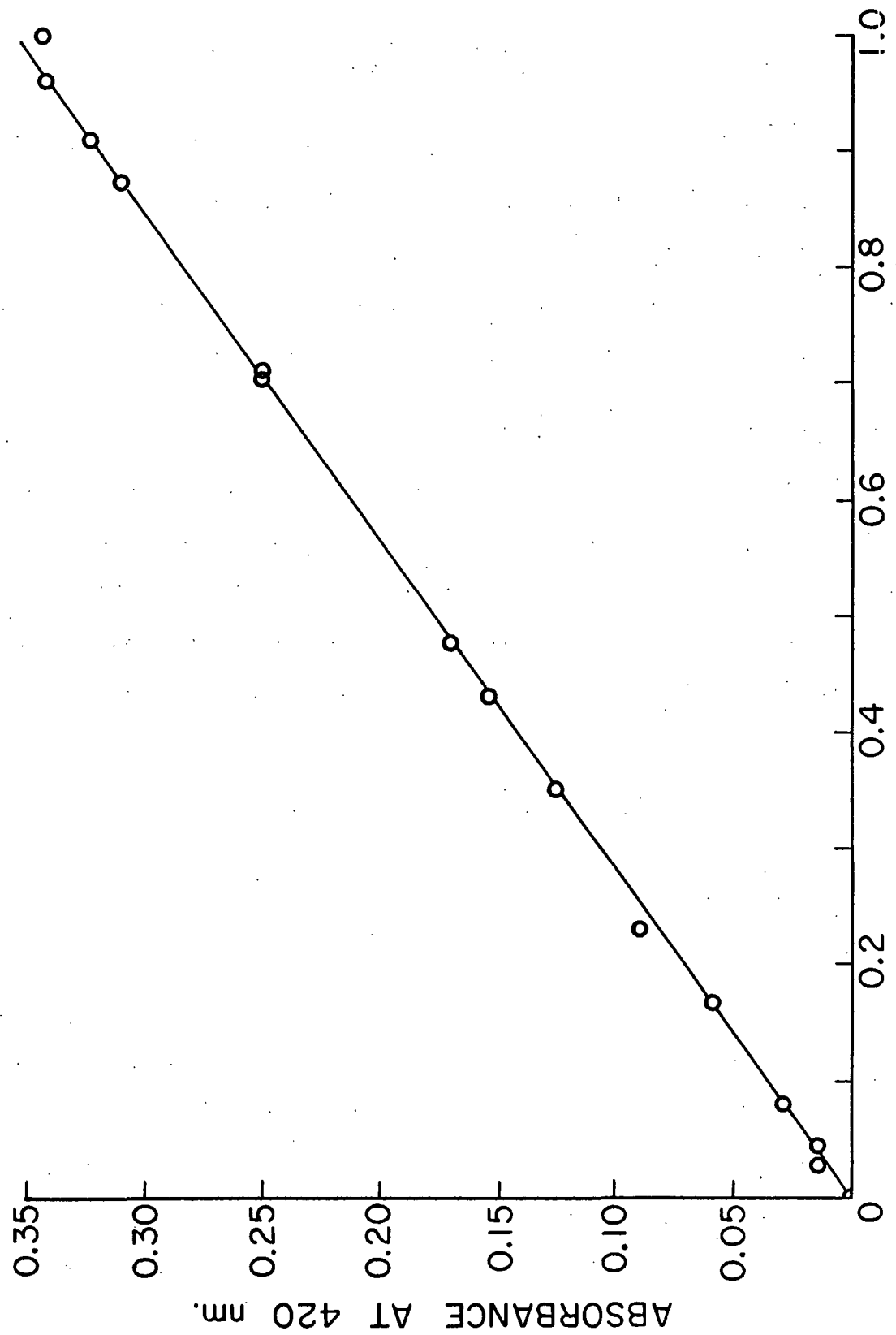

Figure 27. Absorbance at 420 nm. vs. Peroxide Concentration, as Determined by Thiosulfate Titration

and the absorbance at 420 nm. was determined immediately on a Beckman DU Spectrophotometer. Blanks, made up of all the above constituents but the titanium solution, always gave zero absorbance compared with distilled water. So distilled water blanks were generally used in determining absorbances. If the absorbance of a solution increased with time, readings were taken until a maximum value was obtained.

As noted in the text, some peroxide determinations were carried out by a procedure similar to that used by Marklund (47). In this case, less sulfuric acid was used in making up the titanium test solution, to give a final pH of 1-3, instead of ca. 0.

ANALYSIS OF UNREACTED GLUCOSIDE

The concentrations of MBG in reaction samples were determined by a colorimetric phenol-sulfuric acid method (67-69). The sample (1.00 ml.) was diluted to 10 ml., and added to an ion exchange column containing 3 ml. of Amberlite IR-120 (H^+) resin over 3 ml. of Amberlite MB-3 mixed bed resin. It was eluted at 0.5 ml./min., followed by water washes (4 × 7 ml.). All eluent was collected in a 100-ml. volumetric flask, and diluted to the mark. An aliquot of this deionized solution (1.00 ml.) was pipetted into a 10-ml. volumetric flask. Aqueous phenol (2.00 ml., 50 mg. phenol/ml.) was added, followed by rapid addition of 5.00 ml. concd. sulfuric acid. After standing at least 30 minutes, the absorbance at 492 nm. was read against a blank in which water replaced the sugar solution. A Beckman DU Spectrophotometer was again employed. Concentrations of MBG were read from a straight-line calibration curve (Fig. 28), prepared using solutions of known concentrations of MBG or D-glucose.

At least four duplicate determinations were run on each deionized solution. Standard deviations were generally less than 2%.

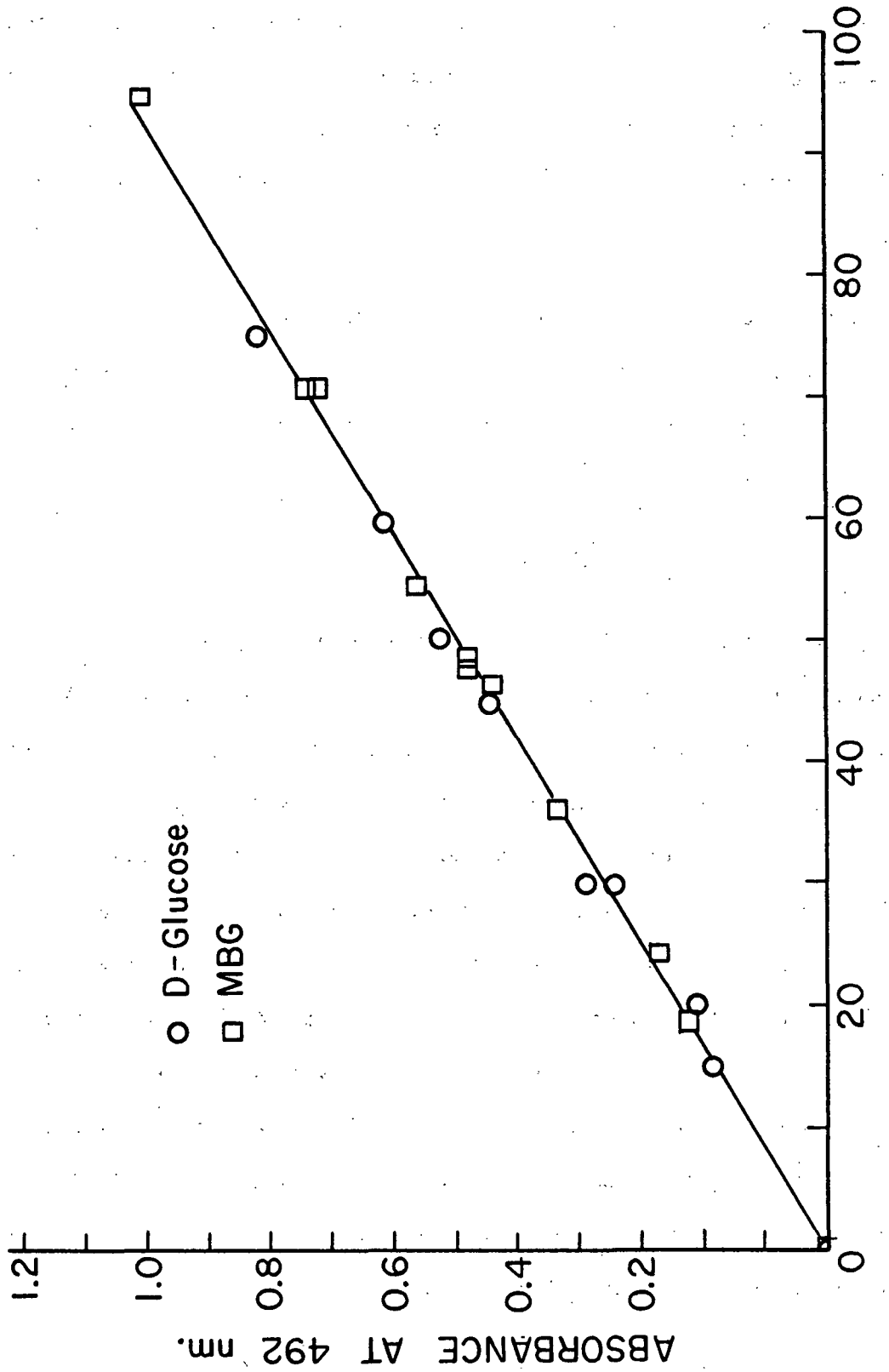

Figure 28. Absorbance at 492 nm. vs. Sugar Concentration

APPENDIX IV

TABULATED DATA

OXYGEN-ALKALI REACTIONS

TABLE IV

METHYL β-D-GLUCOPYRANOSIDE IN 1.25N NaOH AT 120°C.[a]

Reaction	Description	Hr. Reaction 0	0.1	0.25	0.5	1.0	1.5	3.0	5.5	8.0	11.0
75	31 mM MBG; H_2O_2	N.D.[b]	N.D.[b]	N.D.[b]	0.043	N.D.[b]	0.083	N.D.[b]	0.041	0.029	0.026
	75 p.s.i. O_2 Other peroxides	N.D.	N.D.	N.D.	0.017	N.D.	0.016	N.D.	0.175	0.272	0.333
79	31 mM MBG; Unreacted MBG	30.6	N.D.[b]	30.6	30.5	N.D.[b]	29.7	28.7	27.2	25.8	23.6
	75 p.s.i. O_2										
83	31 mM MBG; Unreacted MBG	31.5[b]	N.D.[b]	31.5[b]	31.4	N.D.[b]	31.6	31.4	29.6	27.9	26.0
	25 mM MgO; H_2O_2	N.D.[b]	N.D.[b]	N.D.[b]	0.058	N.D.[b]	0.218	0.317	0.292	0.235	0.229
	75 p.s.i. O_2 Other peroxides	N.D.	N.D.	N.D.	0.002	N.D.	0.006	0.017	0.018	0.061	0.070
86	31 mM MBG; Unreacted MBG	31.7[b]	N.D.[b]	31.2[b]	30.7	N.D.[b]	29.5	28.6	26.7	25.0	23.5
	0.05 mM $FeCl_3$; H_2O_2	N.D.[b]	N.D.[b]	N.D.[b]	0	N.D.[b]	0.007	0	0	0	0
	75 p.s.i. O_2 Other peroxides	N.D.	N.D.	N.D.	0	N.D.	0.036	0.127	0.229	0.307	0.364
92	32 mM MBG; Unreacted MBG	32.3[b]	N.D.[b]	32.3[b]	31.9	31.3	30.3[b]	29.4	27.8	24.7	23.1[b]
	75 p.s.i. O_2 H_2O_2	N.D.[b]	N.D.[b]	N.D.[b]	0.070	0.112	N.D.[b]	0.078	0.059	0.056	N.D.[b]
	Other peroxides	N.D.	N.D.	N.D.	0.014	0.017	N.D.	0.087	0.181	0.254	N.D.
96	32 mM MBG; Unreacted MBG	32.4	31.9	31.3	31.0	N.D.[b]	30.2	29.0	27.2	25.0[b]	N.D.[b]
	0.17 mM H_2O_2; H_2O_2	0.169	0.139	0.098	0.084	N.D.[b]	0.067	0.053	0.039	N.D.[b]	N.D.[b]
	75 p.s.i. O_2 Other peroxides	0	0	0.006	0.008	N.D.	0.028	0.073	0.168	N.D.	N.D.
99	41 mM MBG; Unreacted MBG	41.2	40.5	39.3[b]	38.6	N.D.[b]	37.9	35.2	N.D.[b]	N.D.[b]	N.D.[b]
	0.17 mM H_2O_2; H_2O_2	0.169	0.095	N.D.	0.073	N.D.	0.031	0.019	N.D.	N.D.	N.D.
	75 p.s.i. O_2										
102	31 mM MBG; Unreacted MBG	N.D.[b]	30.9[b]	30.7[b]	30.5	N.D.[b]	29.9	29.0	26.3	24.7	N.D.[b]
	0.075 mM $Na_4P_2O_7$; H_2O_2	N.D.[b]	N.D.[b]	N.D.[b]	0.056	N.D.[b]	0.090	0.070	0.056	0.048	N.D.[b]
	75 p.s.i. O_2 Other peroxides	N.D.	N.D.	N.D.	0.003	N.D.	0.016	0.070	0.151	0.226	N.D.

See end of table for footnotes.

TABLE IV (Continued)

METHYL β-D-GLUCOPYRANOSIDE IN 1.25N NaOH AT 120°C.[a]

Reaction	Description		Hr. Reaction 0	0.25	0.5	1.5	3.0	5.5	8.0	11.0	13.5
105	32 mM MBG; 0.3 mM $K_2C_2O_4$; 75 p.s.i. O_2	Unreacted MBG	32.5	31.9	31.9	30.5	28.8	26.8	25.0	23.3	N.D.[b]
		H_2O_2	N.D.[b]	N.D.[b]	0.070	0.089	0.070	0.056	0.056	0.050	N.D.[b]
		Other peroxides	N.D.[b]	N.D.[b]	0	0.012	0.070	0.154	0.224	0.300	N.D.[b]
107	32 mM MBG; 0.3 mM $K_2C_2O_4$; 0.05 mM $FeCl_3$; 75 p.s.i. O_2	Unreacted MBG	32.6	31.2	30.3	28.9	26.7	24.8	22.8	20.7	N.D.[b]
		H_2O_2	N.D.[b]	N.D.[b]	0.025	0.031	0.022	0.022	0.022	0.020	N.D.[b]
		Other peroxides	N.D.[b]	N.D.[b]	0.003	0.073	0.202	0.359	0.451	0.526	N.D.[b]
116	32 mM MBG; 1.0 mM MgO; 75 p.s.i. O_2	Unreacted MBG	N.D.[b]	32.2	31.7	31.4	31.4	29.0	26.5	24.9	N.D.[b]
		H_2O_2	N.D.[b]	N.D.[b]	0.084	0.268	0.398	0.381	0.336	0.302	N.D.[b]
		Other peroxides	N.D.[b]	N.D.[b]	0.006	0.012	0.008	0.017	0.048	0.059	N.D.[b]
123	32 mM MBG; 95 p.s.i. O_2	Unreacted MBG	31.9	31.8	31.7	30.9	29.6	28.2	25.8	23.9	N.D.[b]
		H_2O_2	N.D.[b]	N.D.[b]	0.042	0.104	0.076	0.061	0.050	0.042	N.D.[b]
		Other peroxides	N.D.[b]	N.D.[b]	0	0.008	0.050	0.115	0.188	0.258	N.D.[b]
125	34 mM MBG; 55 p.s.i. O_2	Unreacted MBG	33.8	33.9	33.8	32.7	31.3	29.4	28.1	26.5	N.D.[b]
		H_2O_2	N.D.[b]	N.D.[b]	0.020	0.061	0.050	0.039	0.025	0.025	N.D.[b]
		Other peroxides	N.D.[b]	N.D.[b]	0.002	0.003	0.017	0.056	0.131	0.174	N.D.[b]
126	32 mM MBG; 1.0 mM MgO; 75 p.s.i. O_2	Unreacted MBG	32.6	32.6	32.4	32.4	31.7	30.2	27.6	25.3	N.D.[b]
		H_2O_2	N.D.[b]	N.D.[b]	0.042	0.240	0.510	0.614	0.546	0.436	N.D.[b]
		Other peroxides	N.D.[b]	N.D.[b]	0.003	0.004	0.005	0.016	0.025	0.043	N.D.[b]
131	32 mM MBG; 1.0 mM MgO; 75 p.s.i. O_2	Unreacted MBG	N.D.[b]	N.D.[b]	32.2	32.1	31.7	29.7	27.7	N.D.[b]	24.0
		H_2O_2	N.D.[b]	N.D.[b]	0.075	0.286	0.544	N.D.[b]	0.464	N.D.[b]	0.328
134	51 mM MBG; 95 p.s.i. O_2	Unreacted MBG	51.2	51.0	50.0	47.7	45.0	N.D.[b]	N.D.[b]	N.D.[b]	N.D.[b]
		H_2O_2	N.D.[b]	N.D.[b]	0.113	0.123	0.084	0.064	0.056	N.D.[b]	0.039
		Other peroxides	N.D.[b]	N.D.[b]	N.D.[b]	N.D.[b]	0.084	0.202	0.305	N.D.[b]	0.485

See end of table for footnotes.

TABLE IV (Continued)

METHYL β-D-GLUCOPYRANOSIDE IN 1.25N NaOH AT 120°C.[a]

Reaction	Description		Hr. Reaction 0	0.25	0.5	1.0	1.5	2.0	3.0	5.0	7.0	10.0
137	30 mM MBG; 0.23 mM H_2O_2; 1.0 mM MgO; 75 p.s.i. O_2	H_2O_2	0.230	0.287	0.344	0.435	0.484	0.477	N.D.[b]	N.D.[b]	N.D.[b]	N.D.[b]
150	31 mM MBG; 1.0 mM KI; 75 p.s.i. O_2	Unreacted MBG	N.D.[b]	31.0	30.8	N.D.[b]	30.4	N.D.[b]	28.7	28.2[c]	27.2[d]	N.D.[b]
		H_2O_2	N.D.[b]	0.022	N.D.[b]	N.D.[b]	0.064	N.D.[b]	0.045	0.042[c]	N.D.[b]	0.028
152	30 mM MBG; 10.0 mM KI; 75 p.s.i. O_2	Unreacted MBG	N.D.[b]	30.0	30.1	29.7	N.D.[b]	29.6[e]	29.5	28.8	28.5	28.2
		H_2O_2	N.D.[b]	0	0	0	N.D.[b]	0[e]	0	0	0	N.D.[b]

[a] All peroxide and glucoside concentrations are expressed in mmole/liter.

[b] N.D. = not determined.

[c] Determined after 4.5 instead of 5.0 hr. reaction.

[d] Determined after 6.2 instead of 7.0 hr. reaction.

[e] Determined after 1.8 instead of 2.0 hr. reaction.

The following table lists reaction rates, in mM/hr., at various glucoside concentrations. These data were obtained from the slopes of plots of "Unreacted Glucoside" vs. "Reaction Time."

TABLE V

EXPERIMENTAL METHYL β-GLUCOSIDE DEGRADATION RATES[a]

Reaction	Description	Glucoside Concentration, mM						
		32.0	31.0	30.0	28.0	26.0	24.0	22.0
79	31 mM MBG; 75 p.s.i. O_2	N.D.[b]	N.D.[b]	0.9	0.7	0.6	0.6	N.D.[b]
83	31 mM MBG; 25 mM MgO; 75 p.s.i. O_2	N.D.[b]	N.D.[b]	0.8	0.7	N.D.[b]	N.D.[b]	N.D.[b]
86	31 mM MBG; 0.05 mM $FeCl_3$; 75 p.s.i. O_2	N.D.[b]	2.0	1.3	0.7	0.6	0.5	N.D.[b]
92	32 mM MBG; 75 p.s.i. O_2	N.D.[b]	1.0	0.9	0.8	0.7	0.6	N.D.[b]
96	32 mM MBG; 0.17 mM H_2O_2; 75 p.s.i. O_2	4.7	1.0	0.8	0.8	N.D.[b]	N.D.[b]	N.D.[b]
99	41 mM MBG; 0.17 mM H_2O_2; 75 p.s.i. O_2	N.D.[b]	N.D.[b]	8.0[c]	1.5[c]	1.2[c]	N.D.[b]	N.D.[b]
102	31 mM MBG; 0.075 mM $Na_4P_2O_7$; 75 p.s.i. O_2	N.D.[b]	N.D.[b]	0.9	0.8	0.7	N.D.[b]	N.D.[b]
105	32 mM MBG; 0.3 mM $K_2C_2O_4$; 75 p.s.i. O_2	N.D.[b]	1.1	1.0	0.8	0.7	0.6	N.D.[b]
107	32 mM MBG; 0.3 mM $K_2C_2O_4$; 0.05 mM $FeCl_3$; 75 p.s.i. O_2	5.0	3.0	2.0	1.2	1.0	0.8	0.7

[a] Expressed in mM/hr.

[b] N.D. = not determined.

[c] Rates for Reaction 99 were determined at 40, 38, and 36 mM MBG instead of 30, 28, and 26 mM.

TABLE V (Continued)

EXPERIMENTAL METHYL β-GLUCOSIDE DEGRADATION RATES[a]

Reaction	Description	Glucoside Concentration mM				
		32.0	31.0	30.0	28.0	26.0
116	32 mM MBG; 1.0 mM MgO; 75 p.s.i. O_2	0.2	1.0	0.9	0.8	0.7
126	32 mM MBG; 1.0 mM MgO; 75 p.s.i. O_2	0.2	1.0	0.9	0.8	0.7
123	32 mM MBG; 95 p.s.i. O_2	N.D.[b]	1.0	1.0	0.8	0.7
125	34 mM MBG; 55 p.s.i. O_2	0.9	0.8	0.8	0.6	N.D.[b]
131	32 mM MBG; 1.0 mM MgO; 75 p.s.i. O_2	0.1	0.9	0.8	0.8	0.7
134	51 mM MBG; 95 p.s.i. O_2	N.D.[b]	N.D.[b]	2.8[c]	2.4[c]	1.9[c]
150	31 mM MBG; 1.0 mM KI; 75 p.s.i. O_2	N.D.[b]	N.D.[b]	0.9	0.8	0.6
152	30 mM MBG; 10.0 mM KI; 75 p.s.i. O_2	N.D.[b]	N.D.[b]	0.2	0.2	N.D.[b]

[a] Expressed in mM/hr.

[b] N.D. = not determined.

[c] Rates for Reaction 134 were determined at 50, 48, and 46 mM MBG instead of 30, 28, and 26 mM.

TABLE VI

PEROXIDE PRODUCED FROM D-GLUCOSE UNDER 75 P.S.I. OXYGEN IN 5% NaOH AT 22°C.
(Cf. Fig. 15)

Time, hr.	Peroxide Concn., mM	Time, hr.	Peroxide Concn., mM
Reaction A 10.0 mM Glucose, 0.1 mM $Na_4P_2O_7$		Reaction B 10.0 mM Glucose, 1.0 mM MgO, 0.1 mM $Na_4P_2O_7$	
0.1	0.087	0.1	0.104
0.25	0.217	0.25	0.227
0.5	0.410	0.5	0.437
1.0	0.740	1.0	0.786
1.5	1.010	1.5	1.047
2.5	1.438	2.5	1.504
4.0	2.058	4.0	2.039
5.5	2.434	5.5	2.460
7.0	2.695		
Reaction C 10.0 mM Glucose, 1.0 mM MgO, 0.05 mM $FeCl_3$		Reaction D 10.0 mM Glucose, 0.05 mM $Fe\ Cl_3$	
0.1	0.088	0.15	0.126
0.25	0.214	0.25	0.227
0.5	0.402	0.5	0.437
1.0	0.735	1.0	0.773
1.5	0.997	1.5	1.000
2.5	1.434	2.5	1.281
4.0	1.881	4.0	1.468
5.0	2.072	5.5	1.538
6.5	2.277	7.0	1.532
Reaction E 3.0 mM Glucose		Reaction F 3.0 mM Glucose, 1.0 mM $MgSO_4$	
0.4	0.038	0.7	0.091
1.6	0.262	1.7	0.304
2.3	0.363	2.3	0.423
3.2	0.457	3.3	0.528
6.0	0.755	5.9	0.803
7.0	0.850	7.4	0.934
8.0	0.930	8.3	1.013

HYDROGEN PEROXIDE STABILITY

TABLE VII

STABILITY OF H_2O_2 AND OF MBG PEROXIDE INTERMEDIATE IN 5% NaOH AT 35°C. IN PRESENCE OF 1.0 mM $MgSO_4$

MBG Reaction Solution		H_2O_2 Added to Same Medium After Original Peroxide Decomposed	
Hours at 35°C.	mM Peroxide	Hours at 35°C.	mM H_2O_2
0	0.097	0	0.086
1.5	0.097	7.0	0.083
5.5	0.095	20.0	0.082
20.6	0.091	24.5	0.081
29.0	0.090		

TABLE VIII

EFFECT OF $MgSO_4$ ON H_2O_2 STABILITY IN 5% NaOH AT 20°C.[a] (Cf. Fig. 13)

$MgSO_4$ Concn., mM	Reaction Time, hr.			
	0.0	22.8	47.0	93.7
0.00	0.424	0.234	0.136	0.049
0.02	0.430	0.308	0.234	0.140
0.04	0.430	0.388	0.353	0.283
0.06	0.430	0.416	0.392	0.336
0.10	0.430	0.424	0.409	0.371
0.20	0.427	0.424	0.416	0.388

[a] Peroxide concentration, in mmole/liter, at the indicated reaction times.

TABLE IX

DECOMPOSITION OF H_2O_2 IN 5% NaOH AT 120°C.
(Cf. Fig. 14)

Time, min.	H_2O_2 Concn., mM	Time, min.	H_2O_2 Concn., mM
Reaction 94 Nitrogen Atmosphere		Reaction 95 30 mM MBG, Nitrogen Atmosphere	
0	0.171	0	0.168
4.0	0.126	4.7	0.029
9.5	0.091	9.7	0.002
13.0	0.075	15.5	0
19.5	0.055	28.5	0
32.5	0.019		
40.5	0.013		
Reaction 129a Nitrogen Atmosphere		Reaction 130 1.0 mM MgO, Nitrogen Atmosphere	
0	0.094	0	0.090
2.8	0.084	3.6	0.090
6.2	0.077	7.3	0.087
10.4	0.068	12.2	0.087
17.2	0.056	19.2	0.086
25.3	0.042	26.5	0.084
34.3	0.031	35.8	0.080
44.9	0.024	45.7	0.079
Reaction 136a 30 mM MBG, 1.0 mM MgO, Nitrogen		Reaction 136b 1.0 mM MgO, Oxygen Atmosphere	
0	0.230	0	0.232
4.0	0.222	3.3	0.231
7.2	0.217	22.2	0.226
14.7	0.203	64.2	0.188
29.7	0.170	162.2	0.129
53.7	0.133	189.7	0.112
90.7	0.088		

TABLE IX (Continued)

DECOMPOSITION OF H_2O_2 IN 5% NaOH AT 120°C.
(Cf. Fig. 14)

Time, min.	H_2O_2 Concn., mM	Time, min.	H_2O_2 Concn., mM
Reaction 136c 30 mM MBG, 1.0 mM MgO, Nitrogen		Reaction 138c 30 mM MBG, 1.0 mM MgO, Nitrogen	
0	0.230	0	0.233
2.8	0.228	3.0	0.231
9.3	0.210	18.0	0.224
19.4	0.187	36.0	0.203
39.8	0.151	66.5	0.170
64.3	0.116	84.0	0.152
92.8	0.091	105.0	0.129
Reaction 138d 1.0 mM MgO, Oxygen Atmosphere		Reaction 139c 30 mM MBG, 1.0 mM MgO, Nitrogen	
0	0.422	0	0.240
3.0	0.421	3.0	0.240
7.1	0.418	15.0	0.224
24.0	0.400	30.0	0.199
45.0	0.368	45.0	0.175
69.0	0.335	60.0	0.155
91.0	0.308	75.0	0.134

POLAROGRAPHIC DATA

TABLE X

POLAROGRAPHIC RESPONSES OF H_2O_2 IN 5% NaOH[a]

Description of Electrolyte Solution	H_2O_2 Concn., mM	i_{max} at -1.1 v., μA.	i_{max} at -1.8 v., μA.
No additives	0.332	1.134	1.770
	(0.145)[b]	0.555	0.790
	0.0559	0.190	0.192
	(0.050)[b]	0.259	0.274
	(0.109)[b]	0.425	0.586
	(0.192)[b]	0.743	1.049
	(0.304)[b]	1.071	1.617
	(0.409)[b]	1.344	2.223
	(0.593)[b]	1.689	3.234
	(0.803)[b]	2.070	4.380
No $FeCl_3$	0.326	1.035	1.650
Ca. 1 mM $FeCl_3$[c]	0	0.114	0.450
Ca. 1 mM $FeCl_3$[c]	0.266	1.878	2.238
Ca. 1 mM $FeCl_3$[c]	0.437	3.130	3.150
Ca. 1 mM $FeCl_3$, ca. 10 mM $MgSO_4$	0.368	0.114	1.80
0.01 mM $FeCl_3$	(2.7)[b]	13.24	14.96
0.01 mM $FeCl_3$, 0.063 mM $MgSO_4$	(2.4)[b]	7.40	12.99
0.01 mM $FeCl_3$, 0.125 mM $MgSO_4$	(2.3)[b]	1.86	12.49
0.01 mM $FeCl_3$, 0.189 mM $MgSO_4$	(2.2)[b]	1.05	12.01
0.05 mM tartrate	(0.184)[b]	0.710	1.010
0.05 mM tartrate, 0.012 mM $MgSO_4$	(0.184)[b]	0.499	1.020
0.05 mM tartrate, 0.038 mM $MgSO_4$	(0.184)[b]	0.152	1.000
0.05 mM tartrate, 0.062 mM $MgSO_4$	(0.184)[b]	0.074	0.990
No $MgSO_4$	(0.05)[b]	0.264	0.292
0.013 mM $MgSO_4$	(0.05)[b]	0.231	0.276
0.038 mM $MgSO_4$	(0.04)[b]	0.076	0.211
0.050 mM $MgSO_4$	(0.04)[b]	0.036	0.200
0.062 mM $MgSO_4$	(0.04)[b]	0.018	0.200

[a] Unless otherwise noted, m = 1.298 mg./sec., t = 5.02 sec., h_{corr} = 60.8 cm., T = 20°C.

[b] Concentrations in parentheses were determined from i_{max} at -1.8 v. (Fig. 17) instead of by direct analysis.

[c] Small waves observed at -0.95 and -1.55 v.

TABLE X (Continued)

POLAROGRAPHIC RESPONSES OF H_2O_2 IN 5% NaOH[a]

Description of Electrolyte Solution	H_2O_2 Concn., mM	i_{max} at -1.1 v., μA.	i_{max} at -1.8 v., μA.
No $MgSO_4$	(0.74)[b]	2.001	4.032
0.012 mM $MgSO_4$	(0.72)[b]	1.410	3.936
0.035 mM $MgSO_4$	(0.71)[b]	0.513	3.864
0.047 mM $MgSO_4$	(0.70)[b]	0.396	3.819
0.058 mM $MgSO_4$	(0.69)[b]	0.309	3.780
8.0 mM tartrate	(0.22)[b]	0.580	1.226
8.0 mM tartrate, 0.012 mM $MgSO_4$	(0.22)[b]	0.490	1.193
8.0 mM tartrate, 0.037 mM $MgSO_4$	(0.21)[b]	0.349	1.156
8.0 mM tartrate, 0.049 mM $MgSO_4$	(0.21)[b]	0.259	1.145
8.0 mM tartrate, 0.061 mM $MgSO_4$	(0.21)[b]	0.192	1.127
No tartrate	(0.27)[b]	0.975	1.456
1.0 mM tartrate	(0.26)[b]	0.883	1.416
4.0 mM tartrate	(0.25)[b]	0.710	1.382
10.0 mM tartrate	(0.25)[b]	0.541	1.353
23.8 mM tartrate	(0.24)[b]	0.320	1.286
No tartrate	(0.26)[b]	1.080	1.404
10.0 mM tartrate	(0.17)[b]	0.450	0.938
10.0 mM tartrate, 1 mM $MgSO_4$	(0.18)[b]	0.070	1.007
30 mM MBG, 1 mM $MgSO_4$	(0.24)[b]	0.023	1.310
No tartrate	0.2434	0.782	1.320
10.0 mM tartrate	0.3008	0.576	1.605
10.0 mM tartrate, 1 mM $MgSO_4$	0.2296	0.358	1.210
30 mM MBG, 1 mM $MgSO_4$	0.2621	0.092	1.388
No additives, h_{corr} = 59.1 cm.	0.410	1.164	2.283
No additives, h_{corr} = 66.4 cm.	0.381	1.110	1.995
No additives, h_{corr} = 48.0 cm.	0.386	1.104	1.719
No additives, h_{corr} = 49.3 cm.	0.452	1.317	2.196
No additives, T = 54°C.	0.3315	2.340	2.820
No additives, T = 37°C.	0.3220	1.662	2.250
No additives, T = 52°C.	0.440	3.273	4.005
No additives, T = 36°C.	0.504	2.589	3.537

[a]Unless otherwise noted, m = 1.298 mg./sec., t = 5.02 sec., h_{corr} = 60.8 cm., T = 20°C.

[b]Concentrations in parentheses were determined from i_{max} at -1.8 v. (Fig. 17) instead of by direct analysis.

TABLE X (Continued)

POLAROGRAPHIC RESPONSES OF H_2O_2 IN 5% NaOH[a]

Description of Electrolyte Solution	H_2O_2 Concn., mM	i_{max} at -1.1 v., μA.	i_{max} at -1.8 v., μA.
0.1 mM $Na_4P_2O_7$	0.406	1.290	2.121
0.1 mM $Na_4P_2O_7$	0.704	1.944	3.957
0 Hr. sample: 30 mM MBG[b] and 1.0 mM $MgSO_4$ in 5% NaOH; H_2O_2 added[b]	0.1241	0.009	0.543
Additional H_2O_2 added to above	0.2679	0.090	1.317
1.1 Hr. sample of above[b]	0.3306	0.150	1.710
Additional H_2O_2 added to 1.1 hr. sample[b]	0.4484	0.252	2.340
3.5 Hr. sample of above[b]	0.724	0.360	4.164
Additional H_2O_2 added to 3.5 hr. sample	0.856	0.405	4.641
0 Hr. sample: 30 mM MBG in 5% NaOH; H_2O_2 added[b]	0.0559	0.090	0.234
Additional H_2O_2 added to 0 hr. sample[b]	0.1102	0.276	0.540
1.3 Hr. sample of above[b]	0.1576	0.419	0.875
Additional H_2O_2 added to 1.3 hr. sample[b]	0.1960	0.486	1.130
4.5 Hr. sample of above[b]	0.1030	0.286	0.460
Additional H_2O_2 added to 4.5 hr. sample	0.1900	0.290	0.910
10 mM glucose	0.105	0.390	0.650
2.3 Hr. sample: 3.0 mM glucose in 5% NaOH[c]	0.363	1.072	2.000
Additional H_2O_2 added to 2.3 hr. sample	1.050	1.840	5.704
7.0 Hr. sample of above[c]	0.850	1.280	4.416
Additional H_2O_2 added to 7.0 hr. sample	1.558	1.816	8.664
2.3 Hr. sample: 3.0 mM glucose and 1.0 mM $MgSO_4$ in 5% NaOH[c]	0.423	0.248	2.436
Additional H_2O_2 added to 2.3 hr. sample[c]	0.958	0.284	5.136
7.4 Hr. sample of above[c]	0.934	0.360	4.944
Additional H_2O_2 added to 7.4 hr. sample	1.612	0.832	9.008

[a] Unless otherwise noted, m = 1.298 mg./sec., t = 5.02 sec., h_{corr} = 60.8 cm., T = 20°C.

[b] Reaction run at 120°C. under 75 p.s.i. O_2.

[c] Reaction run at 22°C. under 75 p.s.i. O_2.

APPENDIX V

DERIVATION OF KINETIC EQUATION FOR METHYL β-GLUCOSIDE DEGRADATION

From Reactions (12) through (21), the steady-stage assumptions can be expressed as follows:

$$d[OOH^-]/dt = k_{13}[G^-][O_2] - k_{14}[OOH^-][M^{+n}] + k_{-14}[\cdot OH][M^{+n+1}] - k_{15}[OOH^-][M^{+n+1}] + k_{-15}[O_2^{\cdot-}][M^{+n}] + k_{18}[O_2^{\cdot-}] + k_{21}[\cdot OH]^2 = 0, \quad (28)$$

$$d[O_2^{\cdot-}]/dt = k_{15}[OOH^-][M^{+n+1}] - k_{-15}[O_2^{\cdot-}][M^{+n}] + k_{17}[R\cdot][O_2] - k_{18}[O_2^{\cdot-}] - k_{19}[O_2^{\cdot-}][M^{+n+1}] = 0, \quad (29)$$

$$d[\cdot OH]/dt = k_{14}[OOH^-][M^{+n}] - k_{-14}[\cdot OH][M^{+n+1}] - k_{16}[\cdot OH][G] + k_{18}[O_2^{\cdot-}] - k_{20}[\cdot OH][M^{+n}] - 2k_{21}[\cdot OH]^2 = 0, \quad (30)$$

$$d[R\cdot]/dt = k_{16}[\cdot OH][G] - k_{17}[R\cdot][O_2] = 0, \quad (31)$$

$$d[M^{+n+1}]/dt = k_{14}[OOH^-][M^{+n}] - k_{-14}[\cdot OH][M^{+n+1}] - k_{15}[OOH^-][M^{+n+1}] + k_{-15}[O_2^{\cdot-}][M^{+n}] - k_{19}[O_2^{\cdot-}][M^{+n+1}] + k_{20}[\cdot OH][M^{+n}] = 0, \quad (32)$$

and $$d[M^{+n}]/dt = -d[M^{+n+1}]/dt = 0. \quad (33)$$

Adding Equations (28) through (33) gives Equation (34):

$$0 = k_{13}[G^-][O_2] - k_{19}[O_2^{\cdot-}][M^{+n+1}] - k_{20}[\cdot OH][M^{+n}] + k_{18}[O_2^{\cdot-}] - k_{21}[\cdot OH]^2. \quad (34)$$

Adding Equations (29), (31), and (32), and subtracting Equation (30):

$$0 = k_{16}[\cdot OH][G] - k_{19}[O_2^{\cdot-}][M^{+n+1}] + k_{20}[\cdot OH][M^{+n}] - k_{18}[O_2^{\cdot-}] + k_{21}[\cdot OH]^2,$$

or

$$[O_2^{\cdot-}] = \frac{k_{16}[\cdot OH][G] + k_{20}[\cdot OH][M^{+n}] + k_{21}[\cdot OH]^2}{k_{19}[M^{+n+1}] + k_{18}}. \quad (35)$$

When (35) is substituted into (34), and terms are collected, the following expression in [•OH] is obtained:

$$0 = 2k_{19}k_{21}[\cdot OH]^2[M^{+n+1}] - [\cdot OH]\{k_{16}[G](k_{18} - k_{19}[M^{+n+1}]) - 2k_{19}k_{20}[M^{+n}][M^{+n+1}]\} - k_{13}[G^-][O_2](k_{18} + k_{19}[M^{+n+1}]). \quad (36)$$

If the quantities a, b, and c are defined as follows:

$$a = k_{20}[M^{+n}]/k_{16},$$

$$b = k_{18}/k_{19}[M^{+n+1}],$$

and

$$c = k_{21}/k_{16}^2,$$

then Equation (36) can be rearranged and simplified to

$$0 = 2c(k_{16}[\cdot OH])^2 - (k_{16}[\cdot OH])\{(b-1)[G] - 2a\} - k_{13}(b+1)[G^-][O_2]. \quad (37)$$

The quantity $\underline{k}_{13}[\underline{G}^-][O_2] = \underline{K}_{12}\underline{k}_{13}[\underline{G}][OH^-][O_2]$ is apparently the rate of hydrogen peroxide production (or rate of glucoside disappearance) during the induction period. From Fig. 3 and 5, the value of this quantity equals ca. 1.7×10^{-4}M/hr., when $[\underline{G}] = 3 \times 10^{-2}$M, $[OH^-] = 1.25$M, and $[O_2] = 75$ p.s.i./H, where H = Henry's law constant. Therefore,

$$\underline{k}_{13}[\underline{G}^-][O_2] = \frac{\underline{K}_{12}\underline{k}_{13}}{\underline{H}}(3 \times 10^{-2})(1.25)(75) = 1.7 \times 10^{-4}\underline{M}/hr.,$$

or

$$\underline{K}_{12}\underline{k}_{13}/\underline{H} = 6.05 \times 10^{-5}.$$

If P_{O_2} is the equivalent partial pressure of oxygen at 25°C., then Equation (37) becomes, at $[OH^-] = 1.25$M:

$$0 = 2c(k_{16}[\cdot OH])^2 - (k_{16}[\cdot OH])\{(b-1)[G] - 2a\} - (7.56 \times 10^{-5})(b+1)P_{O_2}[G]. \quad (38)$$

The overall rate of the reaction is given by

$$d[Z]/dt = k_{13}[G^-][O_2] + k_{16}[\cdot OH][G] = 7.56 \times 10^{-5} P_{O_2}[G] + k_{16}[\cdot OH][G]. \quad (39)$$

Given experimental values of overall rates in 1.25N NaOH, at given levels of P_{O_2} and [G], one can determine values of ($k_{16}[\cdot OH]$) to substitute into Equation (37). Three sets of data are needed to solve this equation in the three unknowns a, b, and c. Table XI lists the sets of data used. Sets A and B were obtained by McCloskey; Set C was obtained in the present research.

TABLE XI

KINETIC DATA USED TO EVALUATE RATE EQUATION IN 5% NaOH

Data Set	[G], M	P_{O_2}, p.s.i.	d[Z]/dt, M/hr.	$k_{13}[G^-][O_2] = 7.56\times10^{-5}P_{O_2}[G]$	$k_{16}[\cdot OH][G] = d[Z]/dt - k_{13}[G^-][O_2]$	$k_{16}[\cdot OH] = k_{16}[\cdot OH][G]/[G]$
A	10^{-2}	75	1.10×10^{-4}	5.67×10^{-5}	5.33×10^{-5}	5.33×10^{-3}
B	10^{-2}	55	8.38×10^{-5}	4.16×10^{-5}	4.22×10^{-5}	4.22×10^{-3}
C	3×10^{-2}	75	9.00×10^{-4}	1.70×10^{-4}	7.30×10^{-4}	2.43×10^{-2}

Substituting these values of $k_{16}[\cdot OH]$ into Equation (38), one obtains from Data Set A:

$$0 = 5.689 \times 10^{-5}c - 1.100 \times 10^{-4}b + 1.066 \times 10^{-2}a - 3.333 \times 10^{-6}, \quad (40)$$

from B:

$$0 = 3.560 \times 10^{-5}c - 8.38 \times 10^{-5}b + 8.44 \times 10^{-3}a + 6.0 \times 10^{-7}, \quad (41)$$

and from C:

$$0 = 1.181 \times 10^{-3}c - 9.00 \times 10^{-4}b + 4.867 \times 10^{-2}a + 5.60 \times 10^{-4}. \quad (42)$$

Solving (40), (41), and (42) simultaneously yields

$$a = 9.35 \times 10^{-2},$$

$$b = 11.19,$$

and $\underline{c} = 4.20.$

Since the solution for (38) is

$$\underline{k}_{16}[\cdot OH] = \frac{\{(\underline{b}-1)[\underline{G}] - 2\underline{a}\} + \sqrt{\{(\underline{b}-1)[\underline{G}] - 2\underline{a}\}^2 + 8(\underline{b}+1)\underline{c}(7.56 \times 10^{-5})\underline{P}_{O_2}[\underline{G}]}}{4\underline{c}}$$

then the rate expression [Equation (39)] at $[OH^-] = 1.25\underline{M}$ becomes

$$\frac{d[Z]}{dt} = 7.56\times10^{-5}P_{O_2}[G]+[G]\left\{\frac{(10.19[G]-0.1870)+\sqrt{(10.19[G]-0.1870)^2+3.084\times10^{-2}P_{O_2}[G]}}{16.8}\right\} \quad (23)$$

The solubility of oxygen, and, hence, Henry's law constant $\underline{H}$, are affected by the concentration of alkali in solution (11,70). Thus, one would have to know the relationship between $[OH^-]$ and $\underline{H} = \underline{P}_{O_2}/[O_2]$, in order to predict rates at other levels of $[OH^-]$.

As noted previously, the first term of Equation (23) ($7.56 \times 10^{-5}\underline{P}_{O_2}[\underline{G}]$) represents glucoside degradation which results from direct oxygen attack ($\underline{k}_{13}[\underline{G}^-][O_2]$). Presumably this is the only significant degradation occurring during the induction period. The remainder of this equation represents degradation resulting from radical attack ($\underline{k}_{16}[\cdot OH][\underline{G}]$). As seen in Table XII, the contribution of the second term, $\underline{k}_{16}[\cdot OH][\underline{G}]$, to the overall rate is much less at lower glucoside concentrations. Thus, when $[\underline{G}] \leq 10^{-2}\underline{M}$, the rate at equilibrium ($\underline{k}_{13}[\underline{G}^-][O_2] + \underline{k}_{16}[\cdot OH][\underline{G}]$) is not much greater than the rate during the induction period ($\underline{k}_{13}[\underline{G}^-][O_2]$). For this reason, an extension of the induction period caused by the addition of magnesium would probably be difficult to detect from kinetic data, at low glucoside concentrations. This could explain McCloskey's observation that magnesium ion had little effect on the degradation of 10 m$\underline{M}$ MBG (11).

Figure 29 allows a comparison between theoretical rates, calculated from Equation (23) (Table XII) and experimental data. The correlation is very good,

TABLE XII

THEORETICAL DEGRADATION RATES OF MBG FROM EQUATION (23)

$[G]$, M	P_{O_2}, p.s.i.	$k_{13}[G^-][O_2]$, M/hr.	$k_{16}[\cdot OH][G]$, M/hr.	$d[Z]/dt$, M/hr.	$\log ([G] \times 10^{-3})$	$\log \frac{d[Z]}{dt} \times 10^5$
5×10^{-3}	35	1.322×10^{-5}	0.547×10^{-5}	1.869×10^{-5}	0.699	0.272
5×10^{-3}	55	2.080×10^{-5}	0.839×10^{-5}	2.919×10^{-5}	0.699	0.464
5×10^{-3}	75	2.837×10^{-5}	1.116×10^{-5}	3.953×10^{-5}	0.699	0.597
5×10^{-3}	95	3.590×10^{-5}	1.371×10^{-5}	4.960×10^{-5}	0.699	0.696
10^{-2}	35	2.640×10^{-5}	2.922×10^{-5}	5.560×10^{-5}	1.000	0.746
10^{-2}	55	4.160×10^{-5}	4.205×10^{-5}	8.370×10^{-5}	1.000	0.923
10^{-2}	75	5.670×10^{-5}	5.290×10^{-5}	1.096×10^{-4}	1.000	1.039
10^{-2}	95	7.190×10^{-5}	6.310×10^{-5}	1.350×10^{-4}	1.000	1.130
1.838×10^{-2}	35	0.486×10^{-4}	1.540×10^{-4}	2.026×10^{-4}	1.264	1.307
1.838×10^{-2}	55	0.764×10^{-4}	1.932×10^{-4}	2.696×10^{-4}	1.264	1.430
1.838×10^{-2}	75	1.041×10^{-4}	2.253×10^{-4}	3.294×10^{-4}	1.264	1.518
1.838×10^{-2}	95	1.320×10^{-4}	2.539×10^{-4}	3.859×10^{-4}	1.264	1.590
3×10^{-2}	35	0.795×10^{-4}	5.976×10^{-4}	6.771×10^{-4}	1.477	1.831
3×10^{-2}	55	1.247×10^{-4}	6.678×10^{-4}	7.925×10^{-4}	1.477	1.899
3×10^{-2}	75	1.700×10^{-4}	7.300×10^{-4}	9.000×10^{-4}	1.477	1.954
3×10^{-2}	95	2.154×10^{-4}	7.833×10^{-4}	9.987×10^{-4}	1.477	1.999
5×10^{-2}	35	0.132×10^{-3}	2.146×10^{-3}	2.278×10^{-3}	1.699	2.357
5×10^{-2}	55	0.208×10^{-3}	2.255×10^{-3}	2.465×10^{-3}	1.699	2.392
5×10^{-2}	75	0.284×10^{-3}	2.355×10^{-3}	2.639×10^{-3}	1.699	2.421
5×10^{-2}	95	0.359×10^{-3}	2.448×10^{-3}	2.809×10^{-3}	1.699	2.448

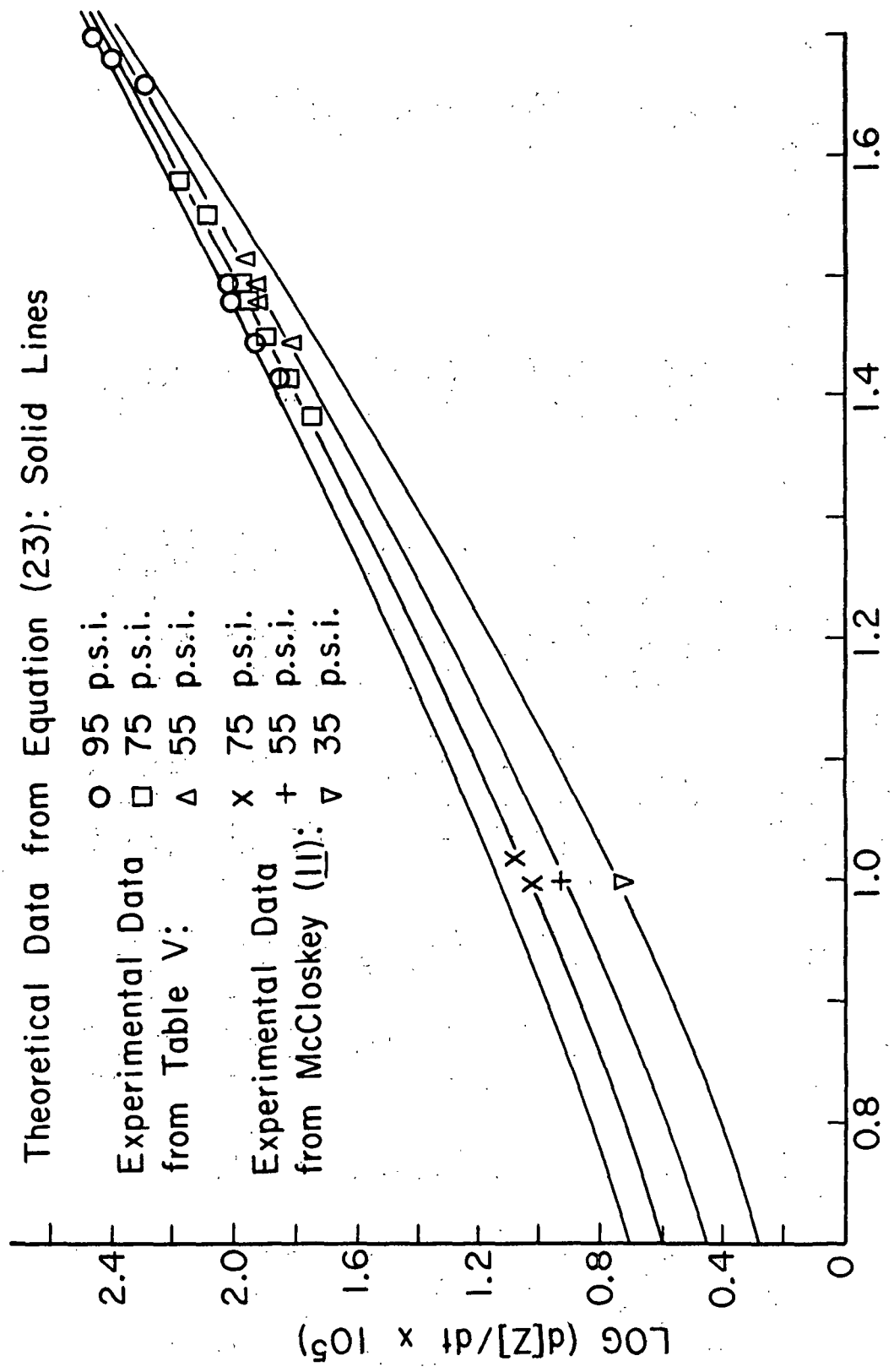

Figure 29. Log MBG Degradation Rate vs. Log Glucoside Concentration

especially when the results of several reaction runs in Table V could be averaged. The reason for the abnormal drop-off of rate in the 50 mM, 95 p.s.i. run is unknown.

The slope of the 75 p.s.i. curve indicates a second-order reaction in glucoside in the range of ca. 14-40 mM MBG. At lower concentrations, the reaction approaches first-order in MBG, as the first term of Equation (23) becomes predominant. Similarly, the oxygen dependency approaches first-order at low glucoside concentrations, and zero-order as [G] increases. Thus, at 50 mM MBG, the effects of oxygen pressure become more difficult to detect.

Hence, Reactions (12) through (21), and the resultant rate Equation (23), seem to describe the MBG degradation quite well. Further research would be needed, however, to firmly establish the validity of this mechanism.